

# LISTERIOSIS RESEARCH

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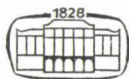
by

Béla RALOVICH

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*LISTERIA MONOCYTOGENES* has fascinated the scientific world since its recognition in the early twenties as a pathogenic microbe infecting both human beings and animals. Recently *Listeria* and its extracts have proved to be valuable tools in experimental immunology. Up-to-date knowledge in the field has been summarized in monographs, and within the past decade three international symposia and a number of national meetings marked the permanent interest in this microorganism and its importance. The discovery of closely related though apparently non-pathogenic organisms of the *Listeria* family not only offered new taxonomic aspects, but also promoted a better understanding of the complex epidemiology and epizootology of this infection, the habitat of the causative organism of *Listeria* infection as well as the conditions which act as promoters in the occurrence of this disease.

The author of the book has been engaged in the study of unknown aspects of *Listeria* and its various actions. He now presents a summary view of this field based upon his own studies and experimental results.



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## ALTERNATIVE PROCEDURES FOR THE GENETIC EVALUATION OF DAIRY SIREs

W. E. VINSON

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Problems of dairy sire evaluation from field recorded data (operating dairy farms) are discussed. A mathematical model is used to describe several factors which must be accounted for if accurate sire evaluations are to result. The relative merits of daughter-dam comparison, contemporary comparison, modified contemporary comparison, and Best Linear Unbiased Prediction sire evaluation procedures are compared.

### Introduction

The accurate genetic evaluation of dairy sires presents several unique and rather complex problems for the animal breeder. For the most part, these problems result directly from the characteristics of the traits for which sire evaluations are desired, and from the dynamic and highly variable nature of the populations which supply the data needed for evaluation. Specifically, the traits for which dairy sire evaluations are desired are generally sex limited and have moderate to low heritabilities. In addition, the large amounts of data required for evaluation must come from operating dairy herds rather than from controlled, experimental farms. Performance is therefore measured in a continuously selected population, dispersed over a wide range of environmental conditions and managerial abilities.

These conditions place some rather stringent requirements on the sire evaluation procedure. The procedure must evaluate sires for traits which they do not themselves express, and for traits in which the differences among sires represent only a small percentage of the total variation in performance. The procedure must accurately estimate these relatively small differences among sires, using performance records which are strongly influenced by variations in climate, housing, feeding, general management, etc. The procedure must account for differences in the intensity with which animals are removed (culled) from the population. An accurate procedure must account for the effects of genetic improvement in the population. The procedure must account for non-random distributions of sires across herds. The procedure must account for the fact that different sires may originate from different populations with differing selection histories, pedigrees, and average genetic values.

In short, an accurate sire evaluation procedure must account for a number of complex biological factors which may be unrelated to a sire's true genetic merit but, nevertheless, have a large effect on the performance of his daughters or other female relatives.

### Methods of sire evaluation

Historically, all methods of dairy sire evaluation have been based on some measure of daughter performance. Female relatives of some type must be used to evaluate sires for sex limited traits expressed only in females. Daughters have been the relatives of choice in artificially bred populations due to their relatively high genetic relationship to the sire, the large numbers which can be produced and measured using artificial insemination (AI), and the wide range of environmental and managerial conditions under which daughters can be performance tested. In effect, the progeny test allows us to "sample" a sire's genotype many times in many different environments.

Several methods of dairy sire evaluation have been used during the last 30 years. These have included,

1. daughter-dam comparisons (DD),
2. contemporary comparisons (CC),
3. modified contemporary comparison (MCC),
4. Best Linear Unbiased Prediction (BLUP).

In order to compare the relative strengths and weaknesses of these various methods it is first necessary to express, in terms of a mathematical model, those factors known to cause variation in the average daughter performances of different sires. The average daughter performance is a basic measure used by all four methods listed above. Any factor, other than the sire's genetic value, which can systematically (i.e. non-randomly) cause the average daughter performance of one sire to differ from that of another, must be accounted for in order to make accurate and unbiased *genetic* comparisons among sires.

The specific mathematical model used depends in part on the nature of the daughter records used for sire evaluation, and on the characteristics of the progeny testing programme. For example, if only first lactation records are used in sire evaluation, the model need not include factors to account for daughter parity, and the effects of daughter culling are greatly reduced. If daughter records are adjusted for age and length of lactation prior to the sire evaluation, the model need not include factors to account for variation in daughter averages caused by these factors.

Supposing sire evaluations are based on first lactation records, adjusted for age at calving and lactation length, a mathematical model for milk yield



which appears useful under Hungarian conditions is as follows:

$$Y_{ijklm} = H_i + G_j + S_{jk} + B_l + e_{ijklm},$$

where

$Y_{ijklm}$  is the first lactation milk yield of the  $m$ th daughter of the  $k$ th sire from the  $j$ th genetic population, having the  $l$ th breeding type of dam, and calving in the  $i$ th herd-year-season,

$H_i$  is the effect on yield of herd-year-season  $i$ ,

$G_j$  is the effect on yield of the average genetic value of sires from genetic group  $j$ ,

$S_{jk}$  is the effect on yield of the transmitting ability of sire  $k$ , expressed relative to the average genetic value of the population from which he originated ( $G_j$ ),

$B_l$  is the effect on yield of the  $l$ th breeding type of dam, and

$e_{ijklm}$  is the effect on yield of random factors which vary among daughters of the same sire, in the same herd-year-season and from the same breeding type of dam.

The term,  $H_i$ , accounts for differences in the quality of environment and level of management which exist in different herds, different years, and different seasons of the year. The presence of this term in the model is to account for the fact that daughters of one sire may occur in far better herds, years, or seasons than daughters of another sire. An accurate sire evaluation procedure will produce evaluations which are *not* functions of the  $H_i$  effects.

In most progeny testing situations, sires cannot properly be considered as arising from a single, common genetic population with a single, common average genetic value. For example, imported and domestically bred bulls will seldom have a common genetic background. In addition, if a population is improving genetically over time, animals (both sires and cows) born in a more recent time period will be, on the average, genetically superior to those born at some distant time in the past. That is, younger sires will have a higher *average* genetic value than older sires. As a numerical example, suppose the average genetic value for milk yield in a population is increasing at the rate of 50 kg per year. Then young sires born in the current year might be expected to be, on the average, 250 kg superior to the average of all bulls born five years ago. The term,  $G_j$ , in the above model allows us to account for this difference, i.e.

$$G_0 = 0 \quad \text{and} \quad G_5 = 250.$$

If the genetic values of one specific old sire and one specific young sire were both 250 kg superior to the averages of their respective populations, their



evaluations are,

$$G_0 + S_{0K} = 250 \quad \text{and} \quad G_A + S_{5K} = 500.$$

Omitting the term,  $G_j$ , from the model is equivalent to assuming that all bulls come from a single population with a single average genetic value. In the illustration, the average of the two genetic values is  $(0 + 250)/2$  or 125 kg, and the evaluations are

$$G_A + S_{0K} = 375 \quad \text{and} \quad G_A + S_{5K} = 375.$$

The result is to overevaluate the old bull and underevaluate the young bull.

As implied in the preceding discussion, the term,  $S_{jk}$ , is the transmitting ability of the  $k$ th sire from the  $j$ th genetic group or population, expressed relative to the average genetic value ( $G_j$ ) of the  $j$ th population.

The term,  $B_i$ , would not appear in a model for U.S. sire evaluation but is necessary under Hungarian conditions. The Hungarian dairy population includes cows of several crossbred breeding types. For example, purebred Holstein bulls may be mated to Hungarian Friesian cows, to  $F_1$  crosses (50% Holstein), to  $R_1$  crosses (75% Holstein), etc. Since the breed composition of a sire's mates may affect the yield of his daughters, it is important to include this factor in the model. If the sire evaluation procedure fails to account for this effect, one sire may appear superior to another simply because a majority of his mates were of a breeding type with high average milk yield.

The final factor in the model ( $e_{ijklm}$ ) is due to differences in all factors not specified, as they affect performance. We assume these factors to be random. For example, we assume that daughters of one sire are treated no better or worse, on the average, than daughters of another sire, within the same herd-year-season, breeding type group.

### Comparisons among methods

Using the mathematical model given above, it is relatively simple to examine the basic measures of performance used in the DD and CC sire evaluation procedures, and to evaluate the modifications to these procedures made by the MCC and BLUP methods. In comparing various methods of sire evaluation, it is important to remember that the goal of sire evaluation is to estimate *only* the transmitting ability of the sire; i.e. the  $G_j + S_{jk}$  terms from the model given previously.

### A) Daughter-dam comparison

In the DD procedure, the performance of each daughter of a sire is compared to that of her dam. The difference in performance is computed for each daughter and dam, and averaged for all daughter-dam pairs,

$$(\bar{Y}_{.jk..} - \bar{Y}'_{.jk..}),$$

where the "prime" denotes performance of the dam and the "bar" indicates an average difference over all herd-year-seasons ( $i$ ), dam breeding types ( $l$ ), and daughters ( $m$ ) of sire  $jk$ . In terms of the factors affecting performance in the previous model, the daughter-dam difference can be written,

$$\begin{aligned} \text{DD} = (\bar{H}_i - \bar{H}'_i) + [(G_j + S_{jk}) - (\bar{G}'_j + \bar{S}'_{jk})] + (\bar{B}_l - \bar{B}'_l) + \\ + (\bar{e}_{ijklm} - \bar{e}'_{ijklm}). \end{aligned}$$

If dams and daughters are treated randomly within herd-year-season and breeding type groups, then

$$E(\bar{e}_{ijklm} - \bar{e}'_{ijklm}) = 0,$$

where  $E()$  denotes "expected value of". Therefore, the basic measure of a sire's performance in the DD procedure will be a function of the following factors:

1. The difference between average herd-year-season effects for daughters and dams ( $\bar{H}_i - \bar{H}'_i$ ). Herds are often the same for dams and daughters, but years and seasons differ. Sires with daughters in the best year-seasons, and mates in the worst year-seasons, are favoured regardless of their true transmitting ability.

2. The difference between the transmitting ability of the sire being evaluated ( $G_j + S_{jk}$ ) and the average transmitting ability for sires of his mates ( $\bar{G}'_j + \bar{S}'_{jk}$ ). Sires mated to cows from genetically inferior sires are favoured, but only partially so, since a part of the genetic inferiority of the dam's sire is transmitted through the dam to the daughter.

3. The difference between the average breeding types of daughters and dams. If the effect of breeding type on milk yield is an exactly proportional one (e.g. rather than additive), this difference will have no effect on the accuracy of DD sire evaluations. The reason is that for a given breed of sire, the breeding type of dams and daughters differs by some constant proportion of the sire's breed.

The major weakness of the DD method of sire evaluation is therefore the failure to adjust for environmental and management differences across

herd-year-seasons. These effects have been shown to account for 30 to 50 per cent of the total differences in milk yield. Since the effect being estimated ( $G_j + S_{jk}$ ) generally accounts for only six to ten per cent of total differences, failure to properly remove herd-year-season differences can seriously bias sire evaluations. This major weakness led to the abandonment of the DD procedure many years ago.

### B) Contemporary comparisons

In the CC procedure, performance of daughters by a particular sire is compared to the average performance of some group of "contemporaries". The definition of contemporaries may vary, and the accuracy of the CC procedure is partly determined by the definition used. As a minimum, contemporaries must be daughters of sires other than the specific one being evaluated, and each contemporary must be of the same lactation as the corresponding daughter.

The goal in defining contemporaries is to designate a group of individuals which is subject to "environmental" conditions nearly identical to those experienced by each specific daughter of a sire. (The term "environmental" as used here refers to any factor other than  $G_j + S_{jk}$ .) By identifying such a group, we can remove differences among sires for these important factors by subtracting average contemporary performance from average daughter performance. The definition of contemporaries therefore generally includes the requirement that contemporaries calve in the same herd, year, and season as the corresponding daughter. Under Hungarian conditions, a more accurate definition would include the requirement that contemporaries be of the same breeding type as the daughter. A potential problem with this definition is that the number of daughters lacking contemporaries, so defined, may be large, and the average number of contemporaries per daughter may be small.

However, for the sake of comparison, suppose that the most accurate definition of contemporaries can be used. That is, suppose contemporaries for a given daughter of a sire are daughters of other sires, calving for the first time in the same herd, year, and season as the given daughter, and of the same breeding type as the daughter. Then the difference between the performance of a daughter and the average performance of her contemporaries is,

$$Y_{ijklm} - \bar{Y}'_{ijk..},$$

which is averaged over all daughters in all herd-year-seasons to give,

$$(\bar{Y}_{.jk..} - \bar{Y}'_{.jk..}),$$



where the "prime" now denotes average performance of contemporaries. In terms of the factors affecting performance, the contemporary difference can be written,

$$CD = (G_j + S_{jk}) - (\bar{G}'_j + \bar{S}'_{jk}) + (\bar{e}_{.jk..} - \bar{e}'_{.jk..}).$$

Note that this difference does not include the effects of herd-year-seasons ( $H_i$ ) or breeding type of dam ( $B_i$ ). It is assumed that their effects on daughters and contemporaries are identical and therefore cancel in the subtraction. If daughters and contemporaries are treated randomly within herd-year-season and breeding type groups,

$$E(\bar{e}_{.jk..} - \bar{e}'_{.jk..}) = 0,$$

and the underlying measure of sire performance in the CC procedure is,

$$CD = (G_j + S_{jk}) - (\bar{G}'_j + \bar{S}'_{jk}).$$

Thus, the contemporary difference is not solely a function of the sire's transmitting ability. It is rather determined by the difference between the transmitting ability of the sire and the average transmitting ability of those (possibly few) bulls who happen to be sires of his daughters' contemporaries.

A major problem in the CC procedure is apparent from the above result. That is, if contemporary sires of one bull happen to be of high average genetic merit, while those of another happen to be of low genetic merit, the contemporary deviation, and therefore the evaluation, of the former will be biased downward with respect to that of the latter. Research in the U.S. (e.g. NORMAN *et al.* 1972) has consistently shown a positive relationship between the genetic merit of a sire, and the average genetic merit of his contemporary sires. That is, if  $(G_j + S_{jk})$  is above average,  $(\bar{G}'_j + \bar{S}'_{jk})$  also tends to be above average. Conversely, if the sire's genetic merit is below average, the average genetic merit of contemporary sires tends to be so also. The reason is that herds with better breeding programmes tend to use *mostly* better than average sires, while herds with poor breeding programmes tend to use *mostly* below average sires. As a result, genetically superior sires tend to be compared with other superior sires, while poorer sires are compared mostly with other poor sires. The CC procedure therefore underevaluates the best available sires and over-evaluates poorer sires.

A second major problem in the CC procedure is the inability to group bulls according to breeding history or pedigree (i.e. according to the average genetic value of the population from which the bull originates). The number of available contemporaries is generally insufficient to require that contemporary sires represent only some specified genetic population. Thus,  $\bar{G}'_j$  may include any combination of genetic group averages, and may vary from



sire to sire. Ignoring this variation is equivalent to assuming that all bulls originate from a single genetic population. For example, Hungarian bulls produced ten years from now will be assumed to be genetically no better than those available today. (If this is true, the national breeding programme is not producing the desired results.) This assumption can lead to serious misranking of bulls. Specifically, under the CC procedure, sires from genetically superior populations are underevaluated relative to those from genetically inferior populations.

A third problem in the CC procedure is its inability to account for genetic improvement in the population. The example given above illustrates an under-evaluation of young bulls due to a failure to properly group bulls. A further discrimination against young bulls occurs in the CC procedure when the population is improving genetically. That is, the genetic value of the contemporaries, themselves, is improving. In the CC procedure, daughters of a young bull are compared to contemporaries from the more recent generations, while daughters of older bulls are compared to contemporaries of earlier generations. Daughters of young bulls are therefore compared to genetically superior contemporaries. As a result, the CC procedure systematically under-evaluates young bulls relative to older bulls.

An additional problem will exist in the CC procedure if it is not possible to require that contemporaries be of the same breeding type as corresponding daughters. If this requirement is impractical, due to small numbers of available contemporaries, the contemporary difference will be,

$$CD = (G_j + S_{jk}) - (\bar{G}'_j + \bar{S}'_{jk}) + (\bar{B}_l - \bar{B}'_l).$$

That is, the evaluation of a sire will be influenced by the breeding type of his mates and the mates of his contemporary sires. Sires mated to breeding types of high yield and compared to contemporaries from lower yielding breeding types will be overevaluated, while sires subject to the reverse situation will be underevaluated.

The deficiencies in the CC procedure cited above produced some very obvious problems with sire evaluations in the U.S. during the mid and late 1960's (EVERETT 1974). Specifically, these problems were:

1. The evaluations of bulls tended to decline with each re-evaluation (the genetic merit of their contemporaries was increasing).
2. Differences between evaluations of the best and worst bulls were smaller than expected (daughters of the best bulls were compared to genetically better contemporaries).
3. Young bulls appeared to be consistently underevaluated relative to older bulls (daughters of young bulls were compared to better contemporaries).
4. Evaluations of highly selected, primarily AI, bulls were only slightly higher than largely unselected, non-AI bulls (the two groups of bulls were

from different genetic populations and were compared to genetically different contemporaries).

These and other problems led to the abandonment of the CC procedure in the U.S. during the early 1970's.

### C) *Modified contemporary comparison*

The MCC procedure is best described as a CC procedure augmented by numerous adjustments which attempt to individually correct the deficiencies of the CC procedure. These adjustments are briefly described as follows (DICKINSON *et al.* 1976):

1. The MCC procedure adds the average genetic merit of contemporary sires to the contemporary deviation of the sire being evaluated. Bulls whose daughters are compared to contemporaries from genetically superior sires thus have a value added to their contemporary deviations, while bulls compared to inferior sires have some value subtracted.

2. Bulls are grouped according to their pedigree index for milk or fat yield, and the average production of cows from sires with similar indexes is used in computing a sire's evaluation. Bulls are therefore not assumed to originate from a single genetic population, with a single average genetic value.

3. Evaluations of all bulls are expressed relative to a single fixed genetic base. Young and old bulls are therefore compared fairly (relative to the same base) even though genetic improvement is occurring in the population.

The MCC procedure appears to be quite an accurate method of sire evaluation (CASSELL *et al.* 1976, CLAY *et al.* 1978, HONNETTE *et al.* 1979). However, since the procedure is essentially a collection of *ad hoc* corrections applied to the CC procedure, its statistical properties are largely unknown and virtually impossible to determine. Most importantly, the MCC procedure can be justified only where extremely large numbers of bulls must be evaluated (e.g. 20 000 to 30 000), and where it is desired to use all lactation records in sire evaluation. Neither of these conditions appear to exist in Hungary.

### D) *Best Linear Unbiased Prediction*

Unlike the MCC procedure, BLUP is not an attempt to correct the CC procedure, but rather is an entirely different method. The derivation of BLUP (e.g. HENDERSON 1972) is somewhat complex and will not be discussed. However, the mixed model equations used in BLUP sire evaluation are quite logical and easy to understand. An example with explanation is given in the Appendix.

A major advantage of BLUP is that its statistical properties are readily described and are desirable in many respects. Some of these properties include:



1. Sire evaluations by BLUP are unbiased. That is, the expected value of a sire's evaluation is his true transmitting ability ( $G_j + S_{jk}$ ).

2. Sire evaluations by BLUP have minimum mean square error (sampling variance) in the class of linear, unbiased predictors of transmitting ability. Sire evaluations with larger sampling errors are more likely to be in serious error.

3. If performance records are normally distributed, sire evaluations by BLUP are maximum likelihood estimates of true transmitting ability.

4. For normally distributed records, sire evaluations by BLUP maximize the probability of correctly ranking sires in a population.

All of the above are highly desirable statistical properties of BLUP. On a more practical level, BLUP procedures correct deficiencies in the CC procedure noted earlier. Variations from sire to sire in the genetic merit of contemporary sires is corrected by simultaneously comparing each sire with *all* other sires, rather than only those who happen to provide contemporaries for his daughters. In the BLUP procedure, sires are grouped according to the genetic population from which they originate. Populations may be defined as deemed appropriate. Bulls may be grouped by country of origin, year of birth, etc. The latter grouping, for example, allows fair comparisons among young and old bulls even though the population is improving genetically. The BLUP procedure is also capable of using relationships among bulls (HENDERSON 1975) when, and to the extent they are known. Use of relationships allows for more precise grouping of bulls (JENSEN 1980) and also yields more accurate evaluations.

In short, BLUP represents the most accurate method of dairy sire evaluation currently available. Further, the procedure seems ideally suited for the large herd, limited sire number and well designed progeny testing programme in Hungary. In addition, the BLUP procedure is extremely versatile and can be adapted to changing conditions in dairy populations and/or computer capabilities. Refinements to BLUP are certain to occur in the future. However, it is unlikely that the basic procedure will ever be replaced as the method of choice for sire evaluation in artificially bred populations.

## Appendix

Suppose we have the following age-adjusted, standard length first lactation records for 15 daughters of four sires in three herd-year-seasons (HYS). Daughters are from two dam breeding types. Sires A and B come from genetic population 1, while C and D are from population 2.



Genetic group	Sire	Mate's breeding type	Herd-Year-Season		
			1	2	3
1	A	1	4100	4200	
		2	4150		5100
	B	1	3600	5150	
		2		5950	
2	C	1	5700		4700
		2	6000	4950	5030
	D	1		4120	
		2		4750	6250

*Record totals*

Herd-Year-Seasons		Genetic groups		Sires		Mate breeding types	
1	23 550	1	32 250	A	17 550	1	31 570
2	29 120	2	41 500	B	14 700	2	42 180
3	21 080			C	26 380		
				D	15 120		

For this example, we will simultaneously estimate the effects (on production) of 11 "factors" — 3 HYS, 2 genetic groups, 4 sires and 2 breeding types. The BLUP mixed model equations for doing so include one equation for each factor. Each equation uses the total yield for the factor and expresses all factors contributing to that total. For example, the equation for HYS 1 is,

$$5H_1 + 3G_1 + 2G_2 + 2S_A + 1S_B + 2S_C + 3B_1 + 2B_2 = 23\,500.$$

The total production for HYS 1 is 23 550 kg. Five records are produced in this HYS and thus the environmental and managerial effect of the HYS is represented 5 times in the total ( $5H_1$ ). Three of the records in HYS 1 are from sires in genetic group 1 ( $3G_1$ ) and 2 from sires in genetic group 2 ( $2G_2$ ). Two records each are by daughters of sires A and C and 1 by a daughter of sire B ( $2S_A + 1S_B + 2S_C$ ). Three daughters in this HYS are from dams of breeding type 1 while 2 are from dams of breeding type 2 ( $3B_1 + 2B_2$ ). No records are from HYS 2 or 3, or from daughters of sire D. Coefficients for these terms are "zero" and these factors are therefore omitted from the equation above. Similarly, the equation for genetic group 1 would be,

$$3H_1 + 3H_2 + 1H_3 + 7G_1 + 4S_A + 3S_B + 4B_1 + 3B_2 = 32\,250.$$

Other equations follow a similar pattern except that the quantity,  $\sigma_e^2/\sigma_s^2$ , is added to the diagonal element of each of the *sire equations*. The reason for doing this is that sires are random variables. That is, we are not solely interested in the performance of the specific daughters included in the evaluation. Rather, we wish to recognize that these daughters represent only a *random sample* of their sire's genetic value. Another similar sized group of daughters may not produce identically, even under identical conditions. Therefore, the ratio of the variance *within* sires ( $\sigma_e^2$  = variance among daughters of the same sire in the same HYS) to the variation *among* sires ( $\sigma_s^2$  = variance among sires) is added to the number of daughters for the given sire. The ratio,  $\sigma_e^2/\sigma_s^2$  is determined by the heritability of the particular trait. For example, if heritability is 0.25,  $\sigma_e^2/\sigma_s^2 = 15$ . The equation for sire A is therefore

$$2H_1 + 1H_2 + 1H_3 + 4G_1 + (4 + 15)S_A + 2B_1 + 2B_2 = 17\,550.$$

for heritability of 0.25. The entire set of 11 equations is given in matrix notation as follows:

$$\begin{bmatrix} 5 & 0 & 0 & 3 & 2 & 2 & 1 & 2 & 0 & 3 & 2 \\ 0 & 6 & 0 & 3 & 3 & 1 & 2 & 1 & 2 & 3 & 3 \\ 0 & 0 & 4 & 1 & 3 & 1 & 0 & 2 & 1 & 1 & 3 \\ 3 & 3 & 1 & 7 & 0 & 4 & 3 & 0 & 0 & 4 & 3 \\ 2 & 3 & 3 & 0 & 8 & 0 & 0 & 5 & 3 & 3 & 5 \\ 2 & 1 & 1 & 4 & 0 & 19 & 0 & 0 & 0 & 2 & 2 \\ 1 & 2 & 0 & 3 & 0 & 0 & 18 & 0 & 0 & 2 & 1 \\ 2 & 1 & 2 & 0 & 5 & 0 & 0 & 20 & 0 & 2 & 3 \\ 0 & 2 & 1 & 0 & 3 & 0 & 0 & 0 & 18 & 1 & 2 \\ 3 & 3 & 1 & 4 & 3 & 2 & 2 & 2 & 1 & 7 & 0 \\ 2 & 3 & 3 & 3 & 5 & 2 & 1 & 3 & 2 & 0 & 8 \end{bmatrix} \begin{bmatrix} H_1 \\ H_2 \\ H_3 \\ G_1 \\ G_2 \\ S_A \\ S_B \\ S_C \\ S_D \\ B_1 \\ B_2 \end{bmatrix} = \begin{bmatrix} 23\,550 \\ 29\,120 \\ 21\,080 \\ 32\,250 \\ 41\,500 \\ 17\,550 \\ 14\,700 \\ 26\,380 \\ 15\,120 \\ 31\,570 \\ 42\,180 \end{bmatrix}$$

There are two dependencies in these equations (sum of rows 1, 2, and 3 equals sum of rows 4 and 5, equals sum of rows 10 and 11) so that an infinite number of solutions exist. To obtain a unique solution, two restrictions are required. The first restriction can be used to establish a genetic base point. All sire evaluations will be relative to this base population. For example, we might choose to set the average value of genetic group 1 as the base or "zero" point by adding the equation,  $G_1 = 0$ , to the 11 above. Similarly, we might define a base breeding type by adding the equation,  $B_1 = 0$ . Solutions using these restrictions are,

$$\begin{array}{llll} H_1 = 4295 & G_1 = 0 & S_A = -61 & B_1 = 0 \\ H_2 = 4328 & G_2 = 392 & S_B = 61 & B_2 = 648 \\ H_3 = 4498 & & S_C = 28 & \\ & & S_D = -28 & \end{array}$$

Sire evaluations are therefore,

$$PD_A = G_1 + S_A = -61$$

$$PD_B = G_1 + S_B = 61$$

$$PD_C = G_2 + S_C = 420$$

$$PD_D = G_2 + S_D = 364$$

In practice, the number of HYS equations can be extremely large. To avoid solving such a large number of equations, HYS equations are absorbed into the remaining equations. Solutions for genetic groups, sires, and breeding types are identical, with or without absorption. However, with absorption, far fewer equations need be solved.

### References

- CASSELL, B. G.—LOSEE, E. A.—NORMAN, H. D.—DICKINSON, F. N. (1976): Stability of proofs for Holstein bulls sampled under limited and multiherd conditions. *J. Dairy Sci.*, **59**, 2095.
- CLAY, J. S.—VINSON, W. E.—WHITE, J. M. (1978): Repeatability as an indicator of stability in contemporary comparison sire evaluations. *J. Dairy Sci.*, **62**, 1132.
- DICKINSON, F. N.—POWELL, R. L.—NORMAN, H. D.—WAITE, L. G.—MCDANIEL, B. J. (1976): The USDA-DHIA modified contemporary comparison sire summary and cow index procedures. USDA-ARS Prod. Res. Rpt. 165.
- EVERETT, R. W. (1974): An extension approach to new sire summaries. *J. Dairy Sci.*, **57**, 972.
- HENDERSON, C. R. (1972): Sire evaluation and genetic trends. Proc. of Animal Breeding and Genetics Symp. in Honor of Dr. J. L. Lush.
- HENDERSON, C. R. (1975): Use of relationships among sires to increase accuracy of sire evaluation. *J. Dairy Sci.*, **58**, 1731.
- HONNETTE, J. E.—VINSON, W. E.—WHITE, J. M.—MCGILLIARD, M. L. (1979): Stability of proofs for high predicted difference Holstein sires. *J. Dairy Sci.*, **62**, 646.
- JENSEN, E. L. (1980): Bull groups and relationships among sires in Best Linear Unbiased Prediction sire evaluation models. *J. Dairy Sci.*, **63**, 2111.
- NORMAN, H. D.—MCDANIEL, B. T.—DICKINSON, F. N. (1972): Regression of daughter and herd mate milk yield on genetic value of herd mate sires. *J. Dairy Sci.*, **55**, 1735.





## QUANTITATIVE CHANGES IN THE INORGANIC AND ORGANIC COMPONENTS OF AMNIOTIC AND ALLANTOIC FLUID, EGG-WHITE AND YOLK DURING INCUBATION OF GOOSE

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Quantitative changes of Na, K, Ca, Cl, P, total protein, albumen, lipid, glucose, cholesterol, creatine, uric acid, carbamide N and  $\text{NH}_3$  were determined in the yolk, the albumen, amniotic and allantoic fluids of incubated goose eggs by referential chemical methods (flame photometry, clinical chemical analyser). Quantitative changes in the organic and inorganic parts were assessed by compartment analysis. It was established that the growing goose embryo consumes mainly carbohydrates initially, followed by proteins and lipids. The presence of amylase and acidic phosphatase was justified in the albumen. The acidic phosphatase content increases from the 21st day of incubation and the amylase content decreases rapidly until the 13th day. The presence of amylase and acidic phosphatase was established in the yolk of goose eggs, too.

In previous experiments (BAINTNER—FEHÉR 1974, FÁNCSI—FEHÉR 1979, FEHÉR—FÁNCSI—MAJOROS 1980, FEHÉR—KÓTAI 1980) we studied the development, histological and ultrastructural changes of chorioallantoic membrane in chickens and geese, and followed the quantitative changes in the contents of amniotic and allantoic fluid, egg-white (albumen) and yolk in the course of incubation (FEHÉR—TELKI—FÁNCSI 1980). However, to acquire a better knowledge of the nutrient uptake and mineral turnover of embryo and foetus and of water circulation within the egg, further basic data were needed. Our investigations were therefore aimed at determining the quantities and changes of Na, K, Ca, Cl, P as well as total protein, albumin, lipid, glucose, cholesterol, creatine, uric acid, carbamide-N and  $\text{NH}_3$  during incubation.

To the question whether there is any relation between the quantitative changes of inorganic and organic matters in amniotic and allantoic fluid, yolk and egg-white in the period of incubation we tried to find an answer by Path analysis, a biometric method replacing the compartmental analysis (EÖRY 1976, JACKEZ 1972).

We obtained valuable information on the flow of materials within the egg during incubation and on the functioning of the embryo's organs.

We have not encountered publications discussing the quantitative changes of inorganic and organic matters in geese. Some authors have studied the quantities of inorganic and organic matters in hen's eggs at different stages of



**Table 1**  
*Methods used in the biochemical laboratory analyses*

Material	Method	Test	Measuring apparatus	Literature
Acid phosphatase	opt. kin. 37 °C, 404 nm	Galeno-pharm. autom.	Satellite clin. chem. aut.	SZÉKELY, L.—BARTALITS, L. (1979)
Amylase	opt. kin. 37 °C, 404 nm	Galeno-pharm. autom.	Satellite clin. chem. aut.	SZÉKELY, L.—BARTALITS, L. (1979)
Cholesterol	sulphosalicylic acid + acetic acid anh. + sulphuric acid recently: cholesterol oxydase	Galeno-pharm. autom.	LKB aut. photom.	SAS, J. (1974)
Total protein	biuret meth.	Galeno-pharm. autom.	Satellite	SZÉKELY, L.—BARTALITS, L. (1979)
Albumin	BCG meth./bromo-creosol green	own	Satellite	SZÉKELY, L.—BARTALITS, L. (1979)
Glucose	glucose oxydase + peroxydase + 4-amino-phenasone	Galeno-pharm.	Satellite	TRINDER (1979)
Na, K, Ca, P, Mg Urea-N NH <sub>3</sub>	flame photometer urease + phenol + hypochloride	Roche test Galeno-pharm	Satellite	Berthelot method (1979)
Creatinine	Jaffé-kinetic	own	Satellite	ZÖLLNER, N.—KISSEL, K. (1979)
Lipid	with phosphoric acid vanilin			
Uric acid	with phosphoric wolfram acid			WENDEL, T. (1979)
Cl	mercury nitrate titration			SCHALES, O. (1979)

incubation (ILJIN 1917, NEEDHAM 1963, ROMANOFF 1960). The total nitrogen content of the allantoic fluid was established by TARGONSKI (1927, cit. NEEDHAM 1963), the residual nitrogen and uric acid contents by NEEDHAM (1963), FISKE—BOYDEN (1926), TARGONSKI (1927) and FRIDERICA (1912, cit. NEEDHAM 1963). The amounts of Na-, K-, SO<sub>4</sub>-, Cl-ions in the yolk, egg-white and chicken embryo were determined by ISEKI (1930, cit. NEEDHAM 1963). The relative quantities of NH<sub>3</sub>, uric acid and carbamide and their changes during incubation of chickens were also pointed out by NEEDHAM (1963). The amount of triglycerids was determined by RIDDLE (1924, cit. NEEDHAM 1963) in the yolk sac and by VLADIMIROV (1926) and SCHMIDT (1929, cit. NEEDHAM 1963) in the blood of the chicken embryo. ROMANOFF (1963)

tabulated the amino acid content of the egg, and the quantities of the individual amino acids. All these data can be used for more information, in spite of the differences between the methods of examination.

### Materials and methods

For the examinations, identical weight (160 g) and shape index eggs of a homogeneous Rhenish stock of geese obtained from the same incubator were used. The sterile method of taking samples of the amniotic and allantoic fluid, thin and thick yolk and egg-white had been elaborated in a preliminary experiment. From the eggs taken carefully out of the incubator, without turning and left unmoved for a minute, the egg-shell and shell membrane above the embryo were cautiously removed. By piercing through the blood vessel deficient area of the allantois with a sterile hypodermic needle, we took allantoic fluid; then, having detached the allantois from it, pierced through the amnion (after the 17th day through the wall of the allantois) to take amniotic fluid. The egg-white is accessible through the egg-shell at the sharp end of the egg. After the removal of egg-shell and shell membrane the yolk was taken out by means of a thick sterile syringe.

Examinations were performed on the 0, 8th, 13th, 18th, 21st and 24th day of incubation. On each of those days samples were collected from 15 eggs. The samples were delivered in ice and immediately processed. The biochemical analysis methods are included in Table 1.

For the biometrical processing of data required in studying the development process, the stochastic compartmental method was chosen. The modelling of our examinations (changes in the organic and inorganic matters of egg-white, amniotic and allantoic fluid, thin and thick yolk as a function of incubation time) called for an analysis technique that made measurable and was able to simulate the given biological process; in our case, the changes in the condition of equilibrium of matters in the egg as within a closed system.

To judge the relations of material equilibrium and analyse the material turnover, the so-called stochastic compartmental model appeared to be a well applicable method. The compartmental analysis is a modelling method that builds up the system studied from a series of separate homogeneous parts (amnion, allantoic fluid, etc.), the so-called "compartments", and is able to disclose connections, causal relations between these parts as for the conditions of material flow.

The compartmental analysis was elaborated to circumscribe conditions of material flow. For the description of changes in the compartments as a function of time, a differential equation system was used.

Modelling with differential equations demands, however, solving a rather complicated system of equations, that requires in any case the use of a computer, and even then the evaluation of the significance level of calculation results remains unsolved.

We made use of the logical analogy called attention to by EÖRY (1976); the compartmental analysis of the problem discussed in our paper has been carried out through the analysis of the time series of standardized partial regression coefficients (Path coefficients), a mathematically simpler method than the one involving the solution of differential equations.

The analogy lies in the fact that the constants of flow, obtained by solving the differential equations, describe the same relations in the graphical representation showing the causal relations as disclosed by the Path coefficients which, however, can be obtained by a simpler calculation. [Further advantage of the Path analysis is that, beside the index expressing the change of the material, those referring to and qualifying other relations can also be arrived at (correlation) coefficient, scatter, T-test, etc.]

Solution by differential equations is indispensable and of great importance when the biological process is simulated on analogous computers with the aid of the compartmental model. In that case, any stage of the process can be simulated.

About the method and mathematical apparatus of the Path analysis applied in our investigations, information can be obtained in the Dictionary of Biometrical Definitions (Biometrical Értelmező Szótár, Mezőgazdasági Kiadó Budapest, 1966) under the respective entries. The actual course of analysis was the following:

1. The relations of the measuring data were analysed with the help of correlation calculations.

2. The correlations were checked for reliability by T-tests.



3. Changes in the material equilibrium as a function of time, action-interaction relations of compartments were studied with the help of Path analysis; scatter was analysed; the type of the best adaptable function was determined; linear, quadratic and cubic components of the intensity factors of flow were pointed out; the significance levels of Path factors checked.

4. The case being an analysis of time series, an autocorrelation test concerning the complex validity of correlations was performed.

In connection with this task we evaluated the autocorrelation of residue and examined the independence on the basis of the average of successive squares of difference and quotient of variance, comparing it with the critical values of Neuman's quotients; reliable correlation can also be accepted only after the repeated control of significance.

## Results

Quantitative changes during incubation in the organic and inorganic matters are shown in figures grouped by the examined components of the egg.

The amount of the *amniotic* fluid is the largest on the 11th day. Its Na and P contents are the highest on the 13th day, while K and Ca contained in it reach maximum on the 18th day (Figs 1, 2). The concentration of Cl increases evenly until hatching. In the period of egg-white swallowing, from the 14th to the 24th day, the amounts of total protein and albumin grow manifold (Figs 3, 4), then suddenly decrease (Fig. 4). The amount of uric acid increases five-fold from the 8th to the 18th day of incubation. The creatinine content rises rapidly until the 13th day, then decreases (Figs 5, 6). The lipid concentration is low up to the 13th day, then increases until hatching, while the concentration of cholesterol becomes gradually lower (Figs 7, 8).

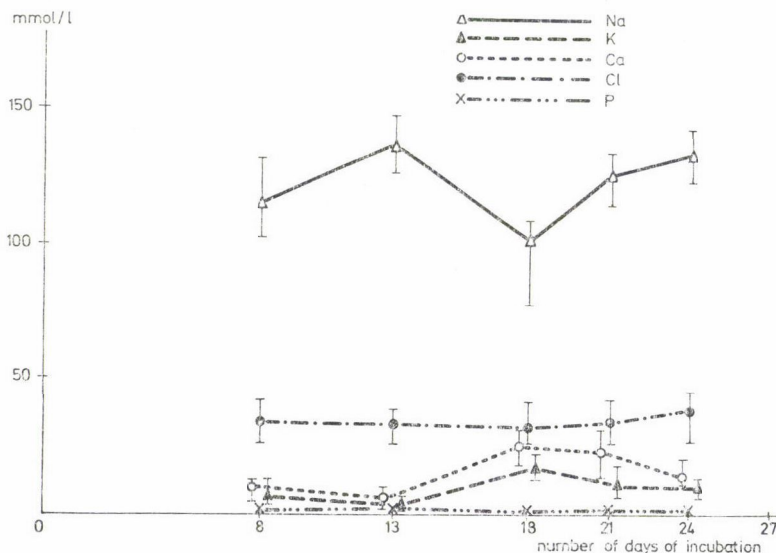


Fig. 1. Amniotic fluid

The Na and Cl contents of *allantois* decrease while its Ca and K contents increase until hatching (Figs 9, 10). The total protein content is at its maximum on the 21st day and decreases afterwards (Figs 11, 12). The albumin, cholesterol

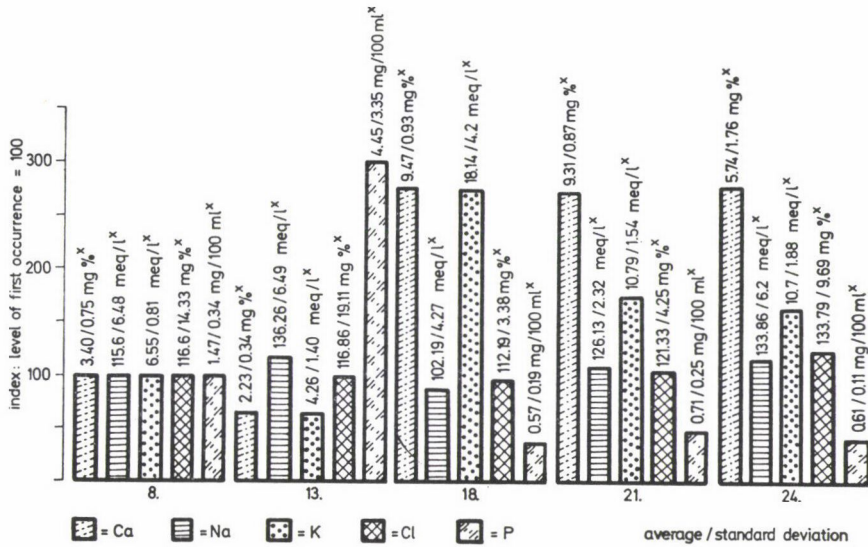


Fig. 2. Quantitative changes in the calcium, sodium, potassium, chlorine and phosphorus contents of the amniotic fluid

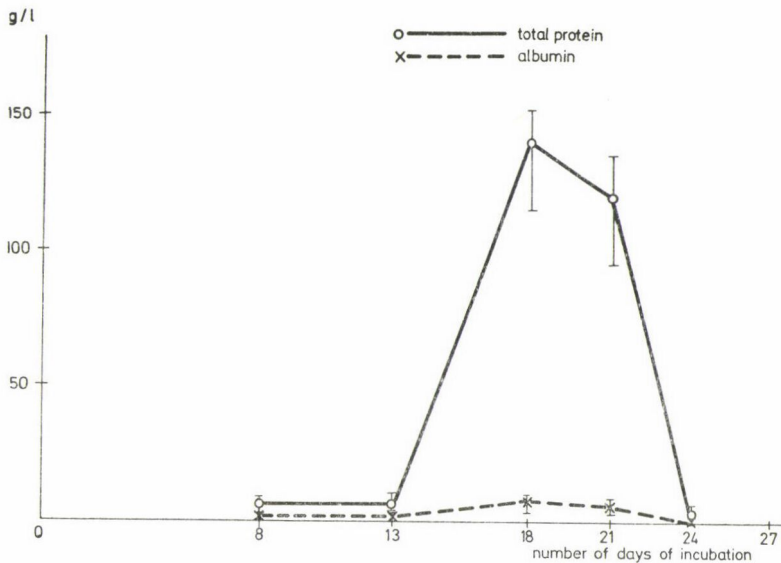


Fig. 3. Amniotic fluid



and urea contents rise until hatching. Intensive increase is found in the creatinine content (Figs 13, 14) after the 18th and in the lipid content (Figs 15, 16) after the 21st day.

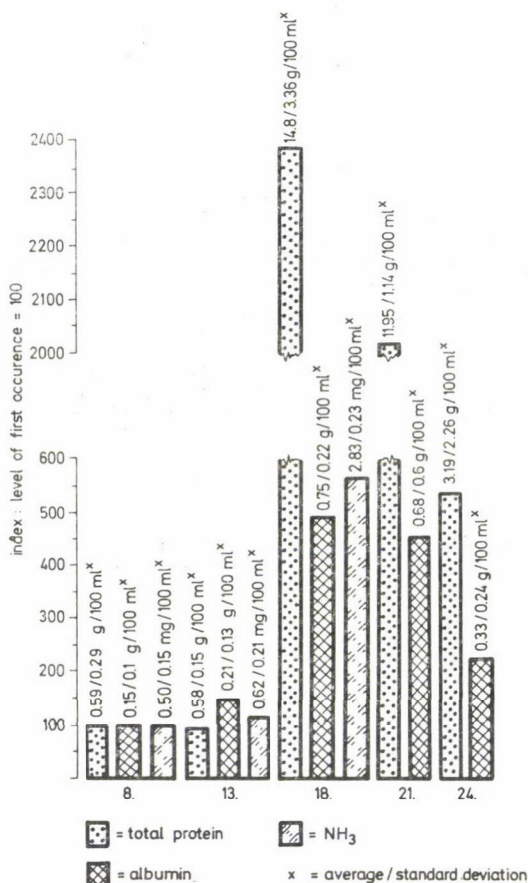


Fig. 4. Quantitative changes in the total protein, albumin and NH<sub>3</sub> contents of the amniotic fluid

In the *yolk sac* the Na content grows threefold in the thick- and more than fivefold in the thin yolk by the 8th day of incubation, then gradually decreases. The K content remains — with slight fluctuations — almost at the same level until hatching. The Ca content is reduced to half by the 8th day, then increases until hatching. The Cl and P contents gradually decrease up to the 13th day, then increase until hatching (Figs 17–20).

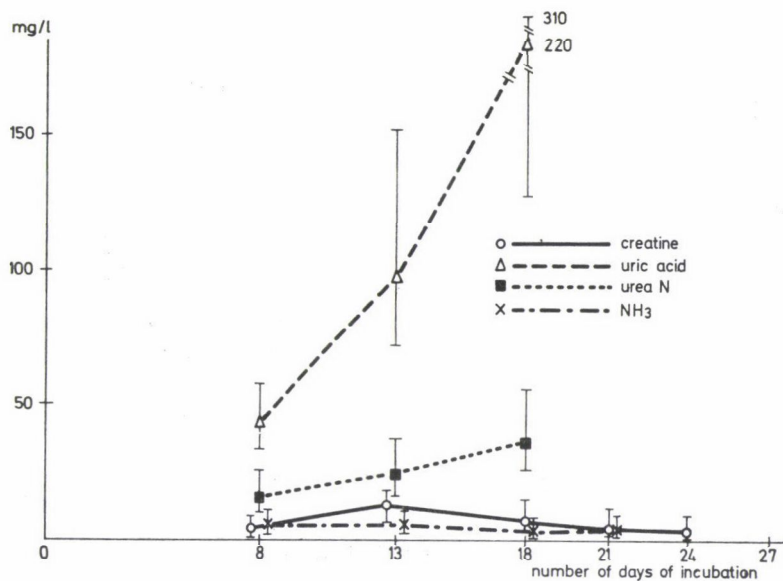


Fig. 5. Amniotic fluid

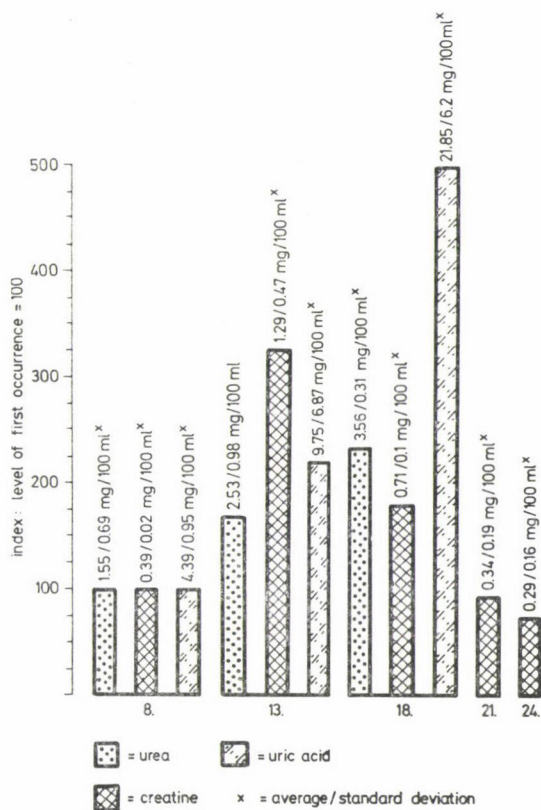


Fig. 6. Quantitative changes in the urea-N, creatine and uric acid contents of the amniotic fluid



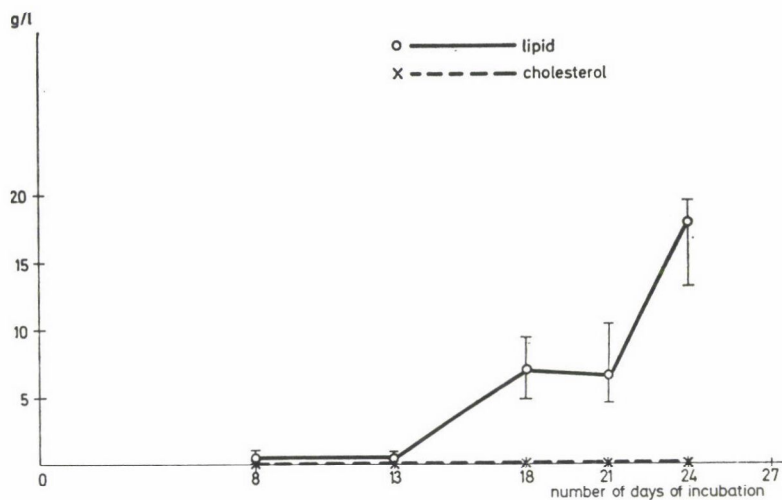


Fig. 7. Amniotic fluid

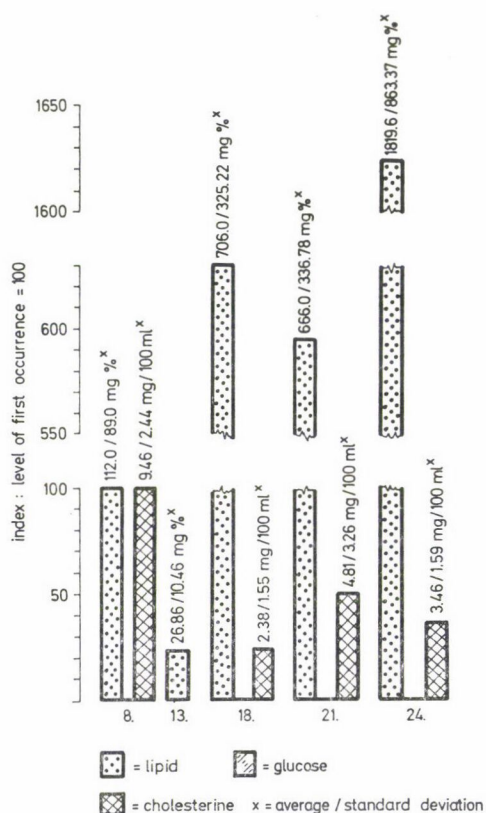


Fig. 8. Quantitative changes in the lipid, cholesterol and glucose contents of the amniotic fluid

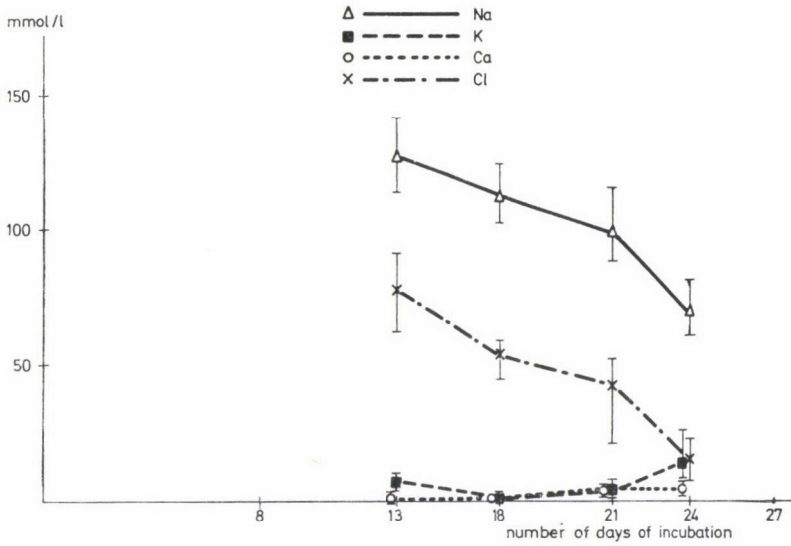


Fig. 9. Allantoic fluid

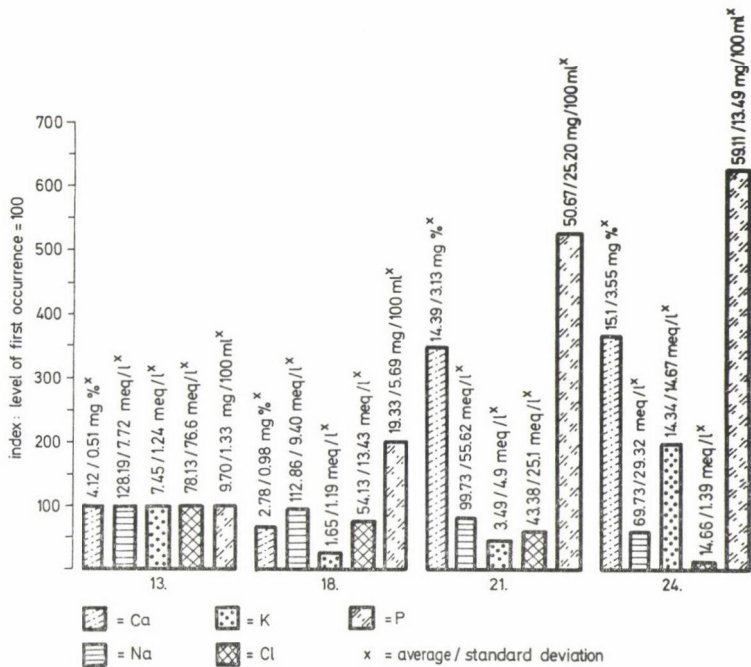


Fig. 10. Quantitative changes in the calcium, sodium, potassium, chlorine and phosphorus contents of the allantoic fluid

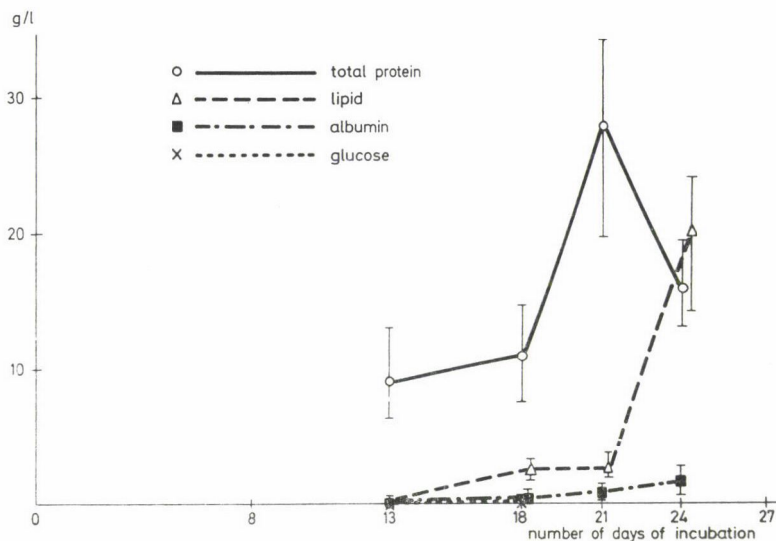
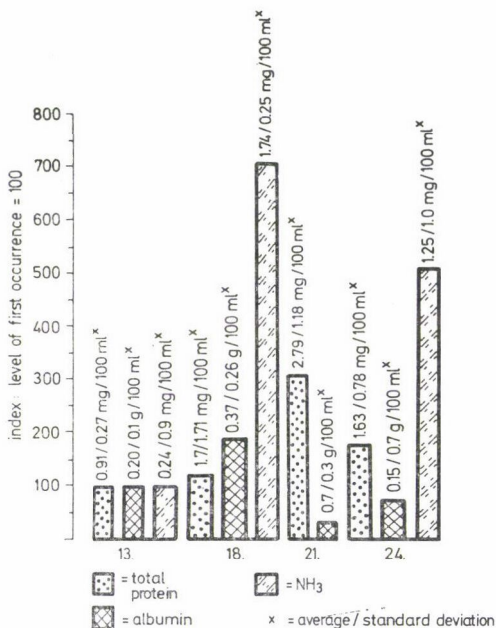


Fig. 11. Allantoic fluid

Fig. 12. Quantitative changes in the total protein, albumin and NH<sub>3</sub> contents of the allantoic fluid



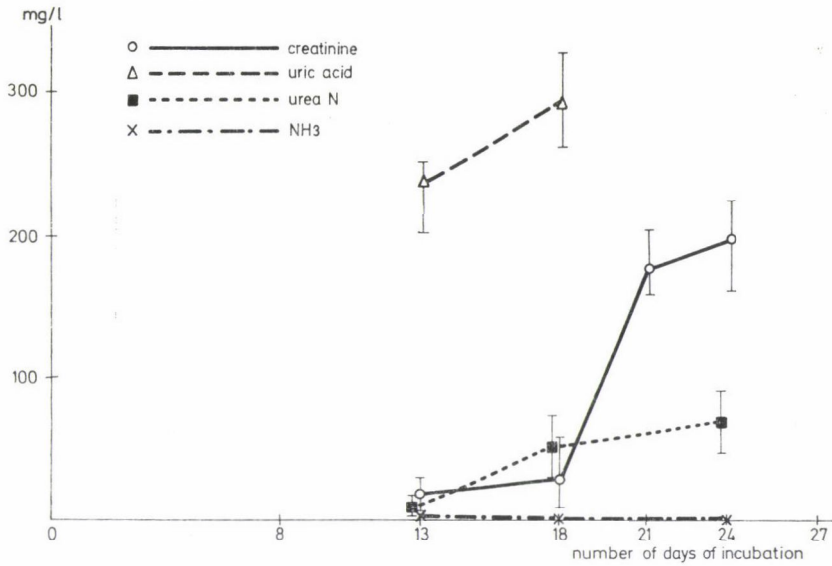


Fig. 13. Allantoic fluid

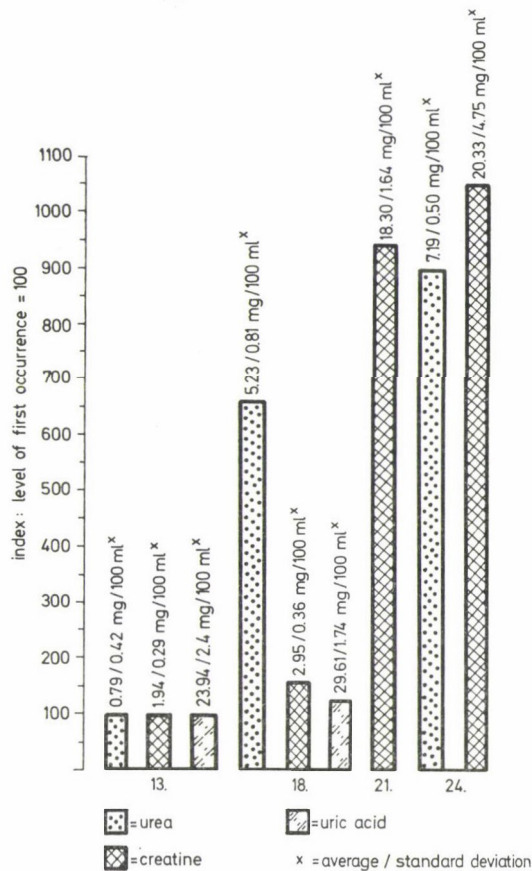


Fig. 14. Quantitative changes in the urea-N, creatine and uric acid contents of the allantoic fluid

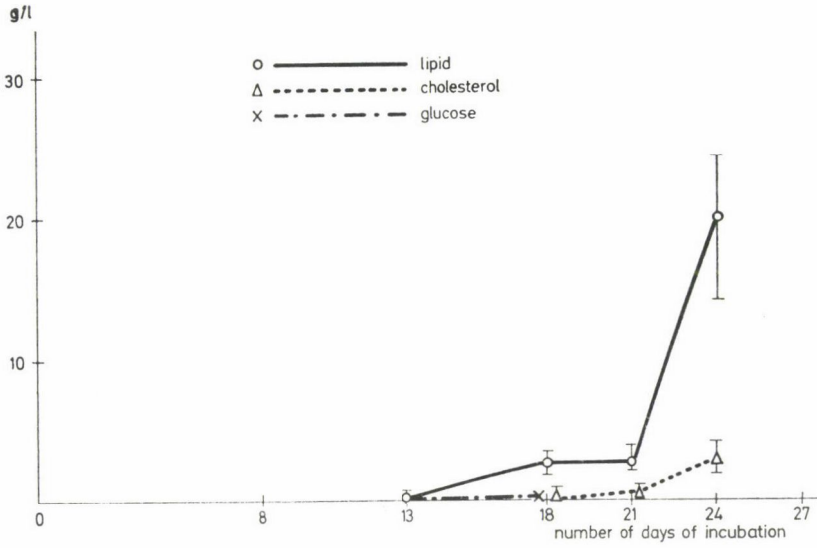


Fig. 15. Allantoic fluid

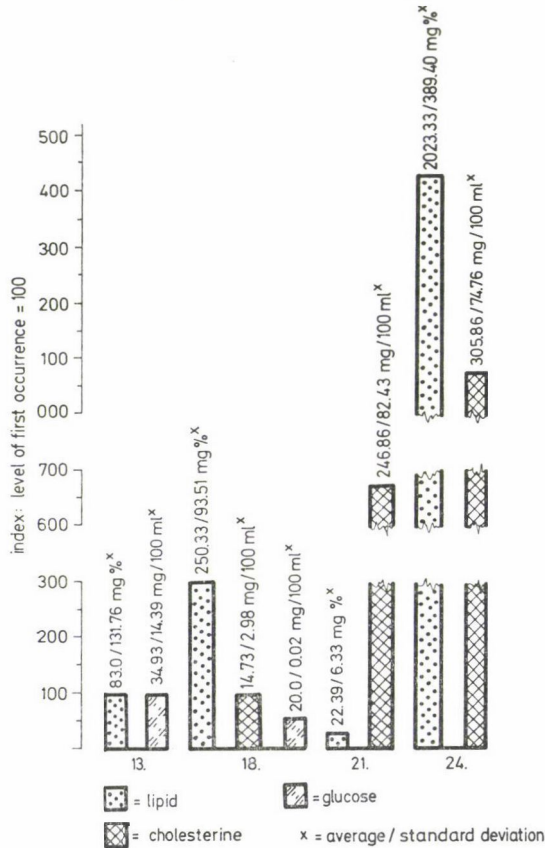


Fig. 16. Quantitative changes in the lipid, cholesterol and glucose contents of the allantoic fluid

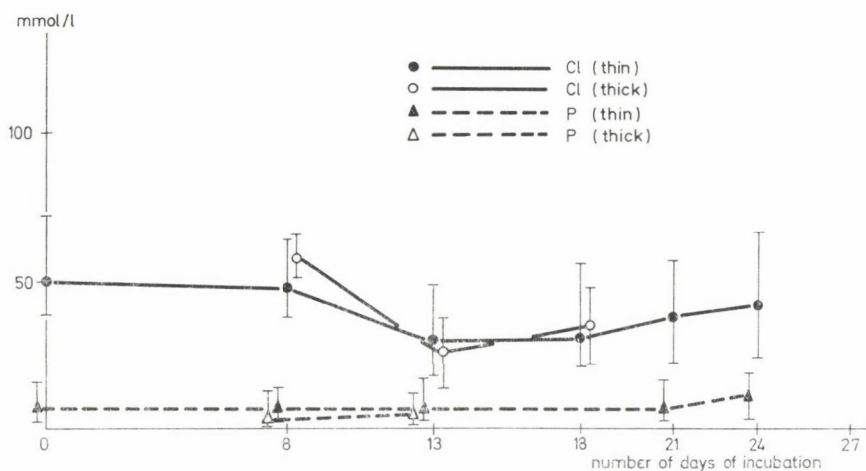


Fig. 17. Yolk

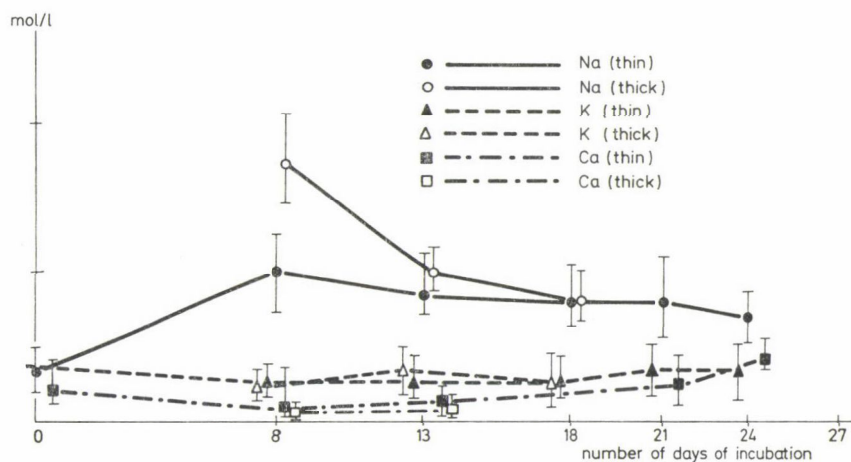
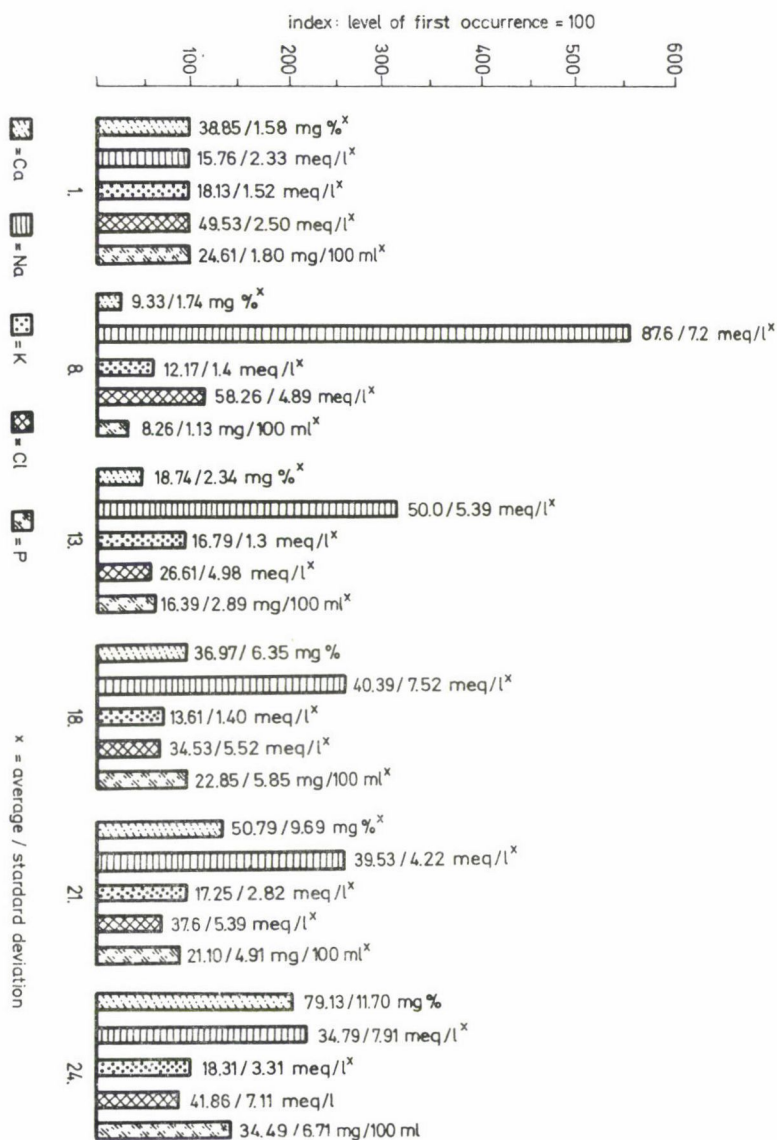


Fig. 18. Yolk

The protein content of the yolk sac hardly changes until the 18th day, then gradually increases till the time of hatching (Figs 21–23). Between the 3rd and 13th day of incubation the total protein content in the thin yolk is high, while in the thick yolk decreases by the 8th day and comes close to the value found in the fresh egg yolk on the 18th day. From then on, the protein concentration shows an intensive growth until hatching (Figs 21, 22). The



Fig. 19. Quantitative changes in the calcium, sodium, potassium, chlorine and phosphorus contents of the thin yolk



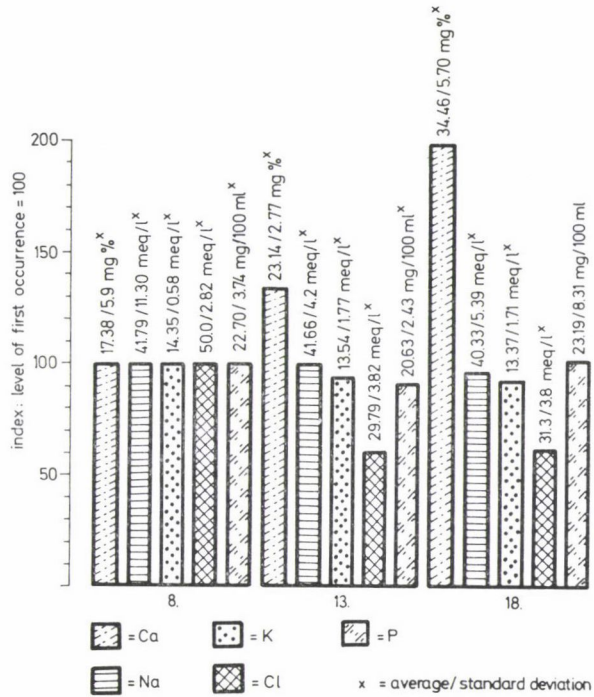


Fig. 20. Quantitative changes in the calcium, sodium, potassium, chlorine and phosphorus contents of the thick yolk

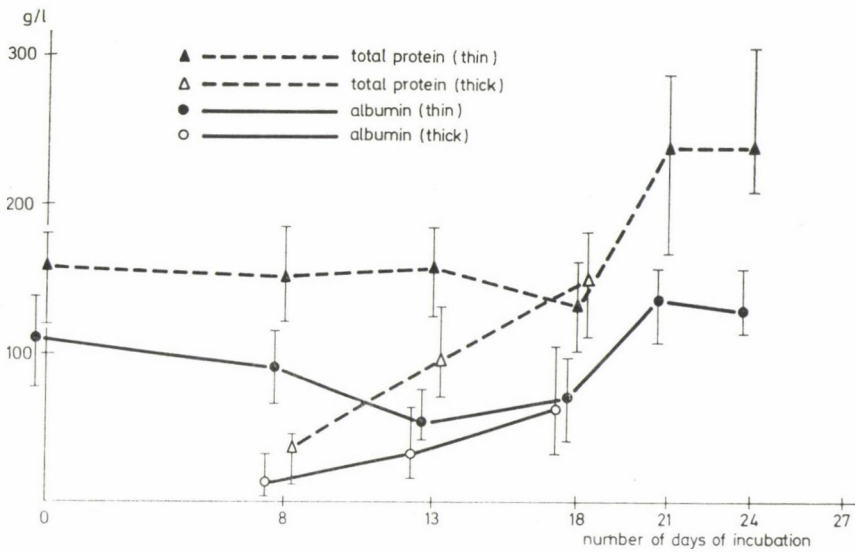


Fig. 21. Yolk

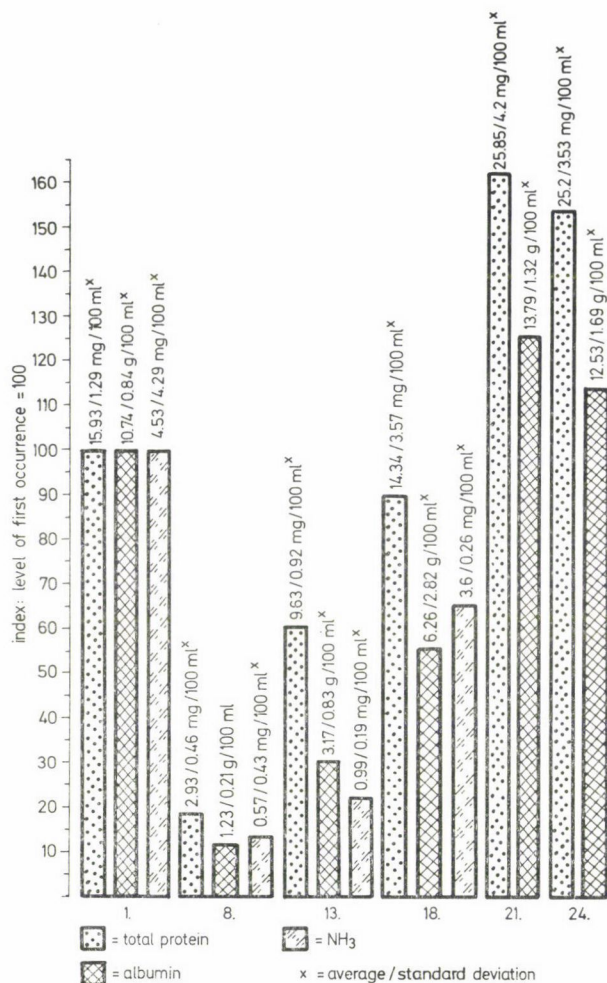


Fig. 22. Quantitative changes in the total protein, albumin and NH<sub>3</sub> contents of the thin yolk

glucose content of the yolk sac is low and decreases rapidly from the 8th to the 18th day (Fig. 24). The lipid content of the thin yolk hardly changes until the 18th day of incubation, then suddenly falls; in the thick yolk it rises by 80 per cent by the 8th day while on the 13th day is lower than in the thin yolk (Figs 25–27). The cholesterol content as well as the concentration of creatinine in the yolk sac decreases until hatching. The urea content of the thin yolk increases fourfold by the 8th day, then suddenly drops. The concentration of uric acid decreases until the 13th day in the thin yolk and increases between the 3rd and 18th day in the thick yolk (Figs 28–30).



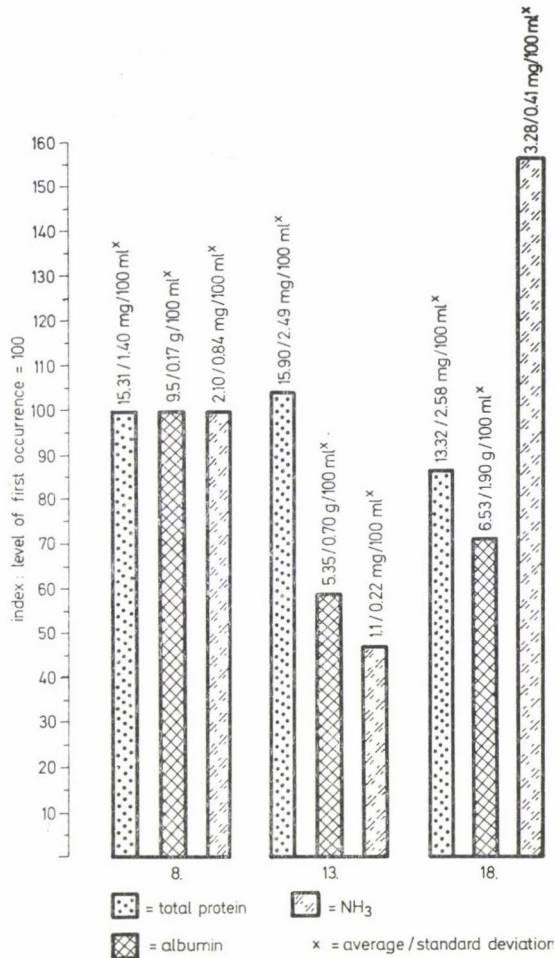


Fig. 23. Quantitative changes in the total protein, albumin and NH<sub>3</sub> contents of the thick yolk

The amylase activity is highest at the beginning of incubation, then gradually decreases. The activity of acid phosphatase remains for a long time relatively low, but after the 21st day increases fourfold (Figs 31, 32, 33).

The Na content of the egg-white increases until the 18th day, its Cl content until the 13th day, then both decrease. The K content becomes slightly lower by the 18th day, then after the 21st day of incubation suddenly rises twice as high as before. From the 18th day the Ca content increases to some extent, the P content remains low and hardly changes until hatching (Fig. 34).

The total protein content is reduced to less than one-third by the 8th day, remains unchanged until the 18th day, then suddenly rises. The albumen slightly increases until the 8th day, then more or less maintains its level until hatching (Fig. 35). Glucose is present in small quantities. The lipid content becomes remarkably high by the 21st day (Fig. 37); the amounts of urea and

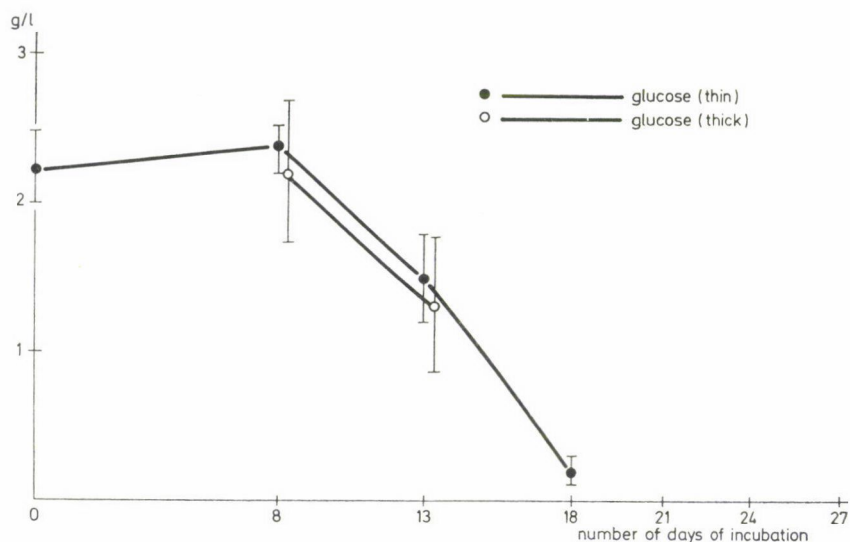


Fig. 24. Yolk

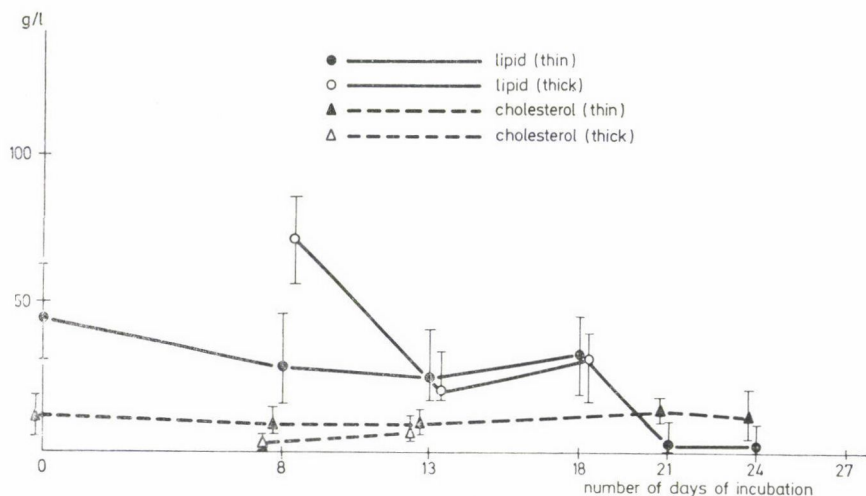


Fig. 25. Yolk

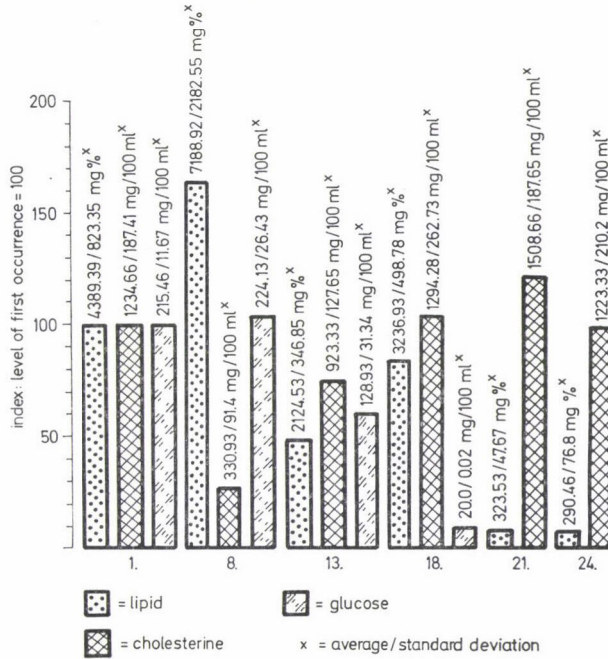


Fig. 26. Quantitative changes in the lipid, cholesterol and glucose contents of the thin yolk

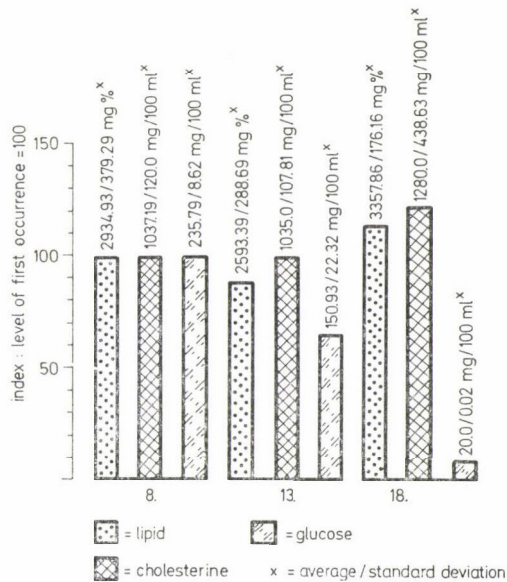


Fig. 27. Quantitative changes in the lipid, cholesterol and glucose contents of the thick yolk



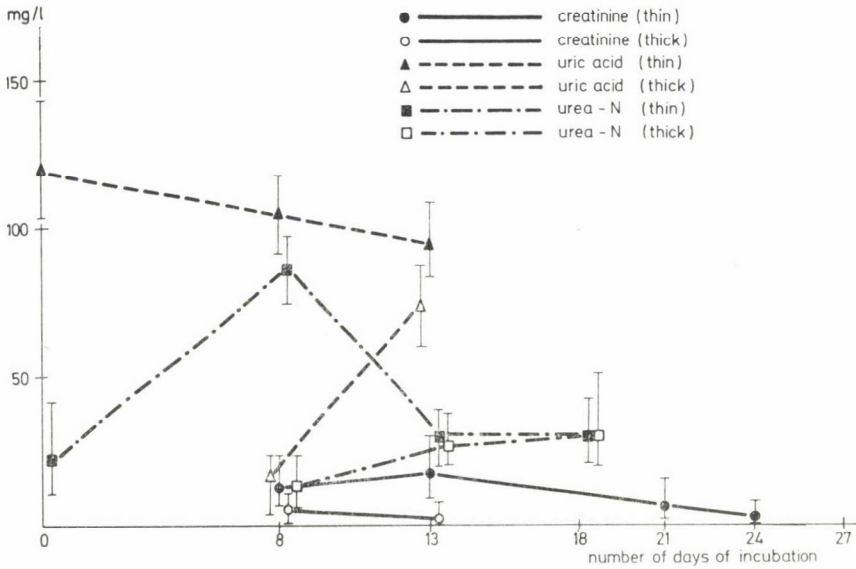


Fig. 28. Yolk

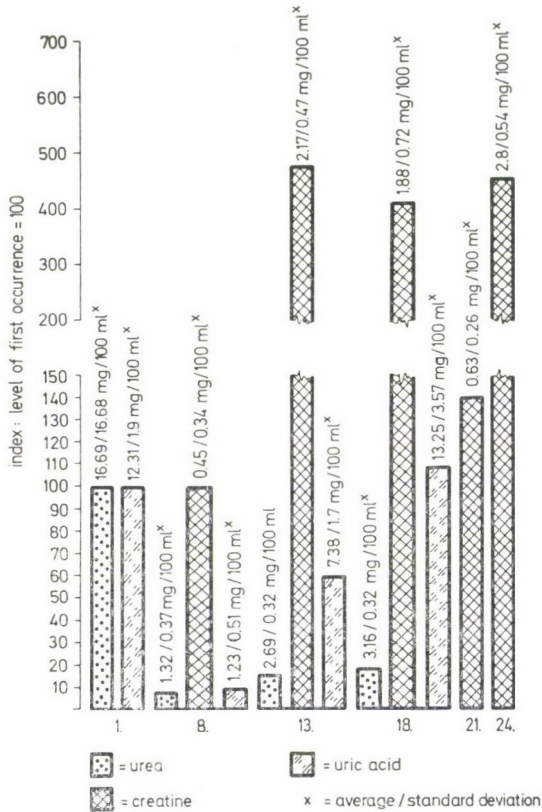


Fig. 29. Quantitative changes in the urea-N, creatine and uric acid contents of the thin yolk

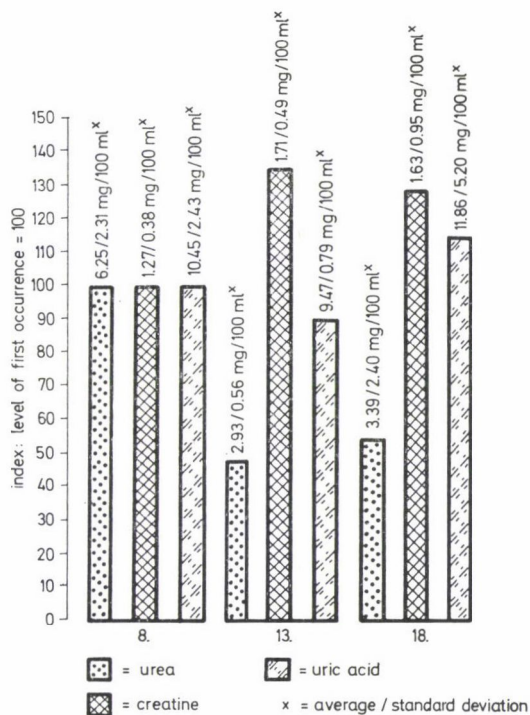


Fig. 30. Quantitative changes in the urea-N, creatine and uric acid contents of the thick yolk

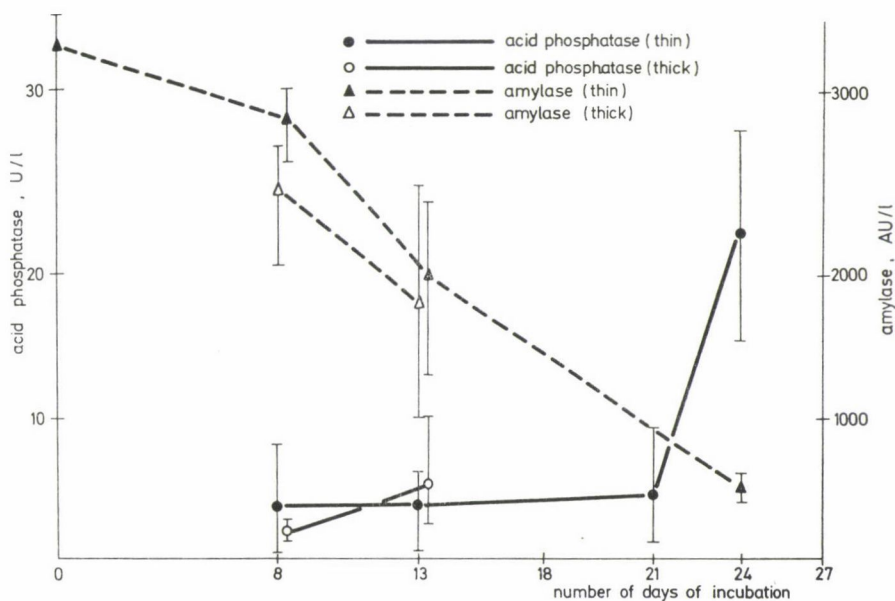


Fig. 31. Yolk

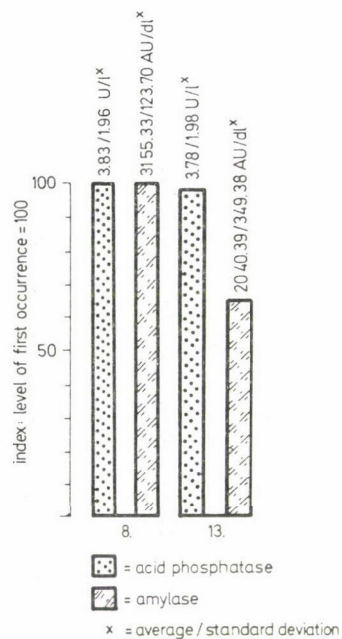


Fig. 32. Changes of acid phosphatase and amylase activity in the thick yolk

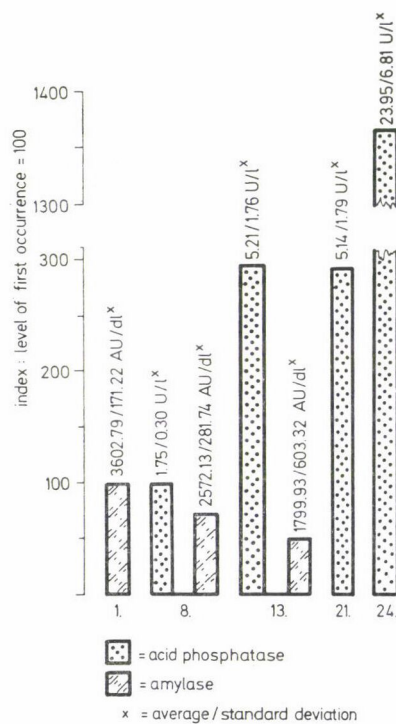


Fig. 33. Changes of acid phosphatase and amylase activity in the thin yolk



uric acid increase only until the 18th day (Fig. 36). The amylase activity is very high until the 8th day of incubation, then lessens; the activity of acid phosphatase increases suddenly after the 21st day (Fig. 38).

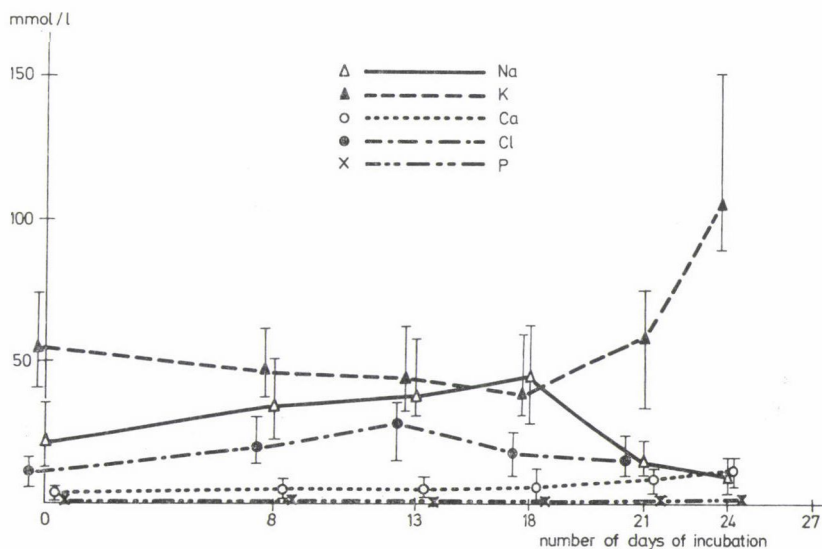


Fig. 34. Egg-white

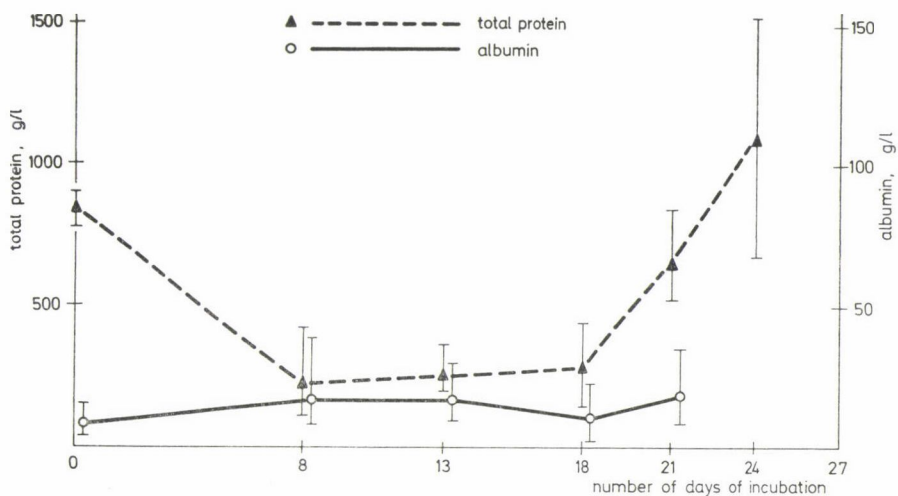


Fig. 35. Egg-white

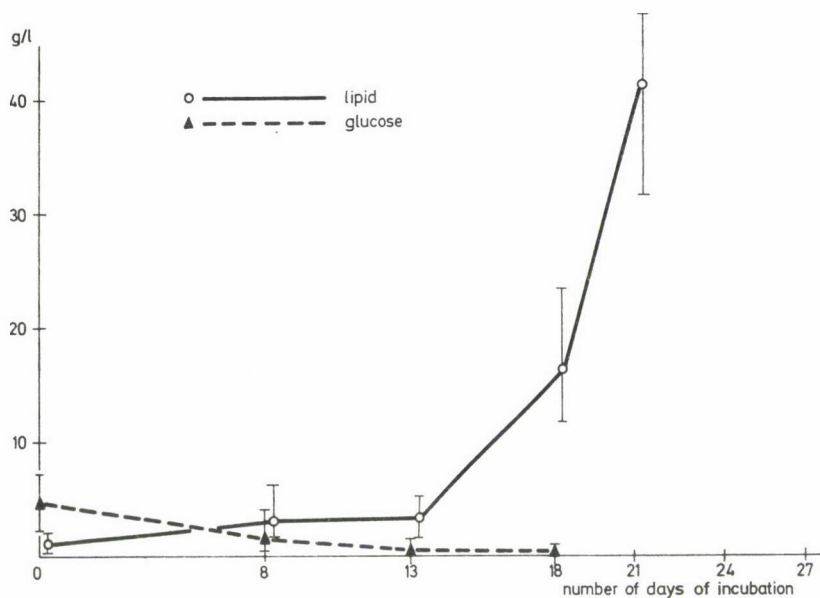


Fig. 36. Egg-white

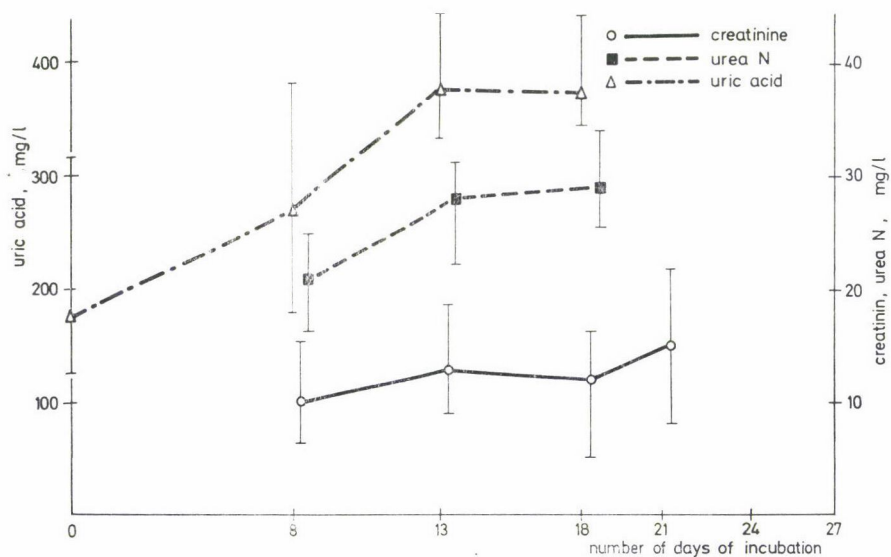


Fig. 37. Egg-white

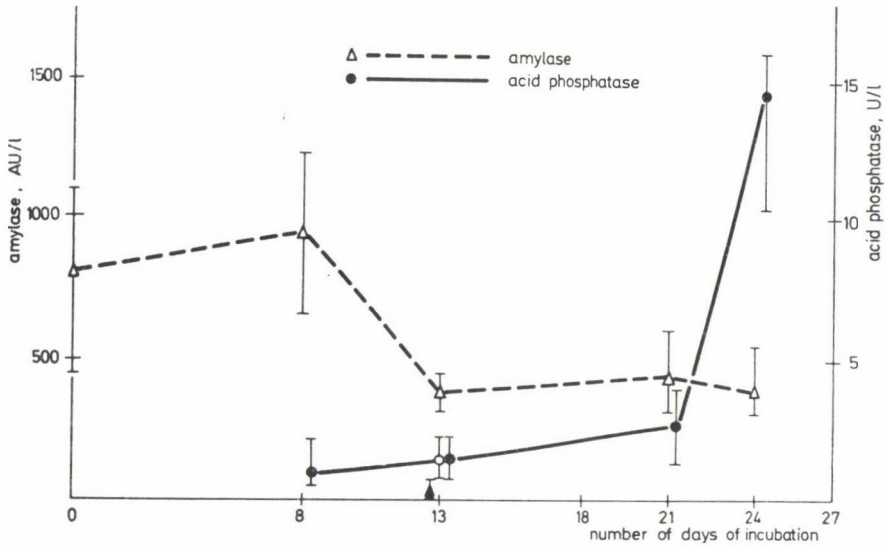


Fig. 38. Egg-white

### Mathematical analysis

In the stochastic analysis two components at a time were examined, for possible correlation between changes in the same organic or inorganic matter contained in them:

There is no correlation between the changes of *total protein contents in the amniotic and allantoic fluid*. The coinciding increase of albumin concentration in the two fluids is accounted for by the Path analysis, and the relationship is not disturbed by autocorrelation. The *lipid content* from the 13th day on suggests a relationship similar to the former ones, by the dynamics of change does not make the relationship acceptable, and the latter is disturbed by the close autocorrelation, too. The cholesterol content shows an increase in the allantoic and a parallel decrease in the amniotic fluid. As a function of time, high (95%) significance can be pointed out, though its value is somewhat reduced by the autocorrelation. The parallel increase in the *urea nitrogen* contents of the two fluids during incubation is significant. The gradual increase of uric acid in the two fluids between the 13th and 18th day of incubation shows a close correlation not disturbed by autocorrelation. With the steady decrease of *Na contents* in the two fluids, the significance is 95%, though autocorrelation slightly reduces it. The increase of *Cl content* in the amniotic fluid and its parallel decrease in the allantoic fluid are in significant correlation only from the 13th to the 18th day, and no longer afterwards.



On the basis of the Path analysis, the increase in the *total protein content of the amniotic fluid* and the decrease in the *protein content of egg-white* between the 8th and 18th day of incubation are correlated. It is explained by the protein inflow in the period between the 13th and 18th day. The higher extreme values are due to the periodical nature of *egg-white* swallowing by the embryo. The gradual increase in the albumen content and decrease in the  $\text{NH}_3$  content of either of the amniotic fluid and egg-white are correlated and significant not even disturbed by the autocorrelation. The *urea nitrogen* contents of the two components increase parallel until the 18th day. The path index is 0.89, autocorrelation does not exist. The  $\text{NH}_3$  content increases until the 18th day, then decreases in the egg-white; and decreases until the 18th day then rises in the amniotic fluid. This correlation is significant; changes in time are correlated, autocorrelation is not present. The *Cl content* of the amniotic fluid slowly but steadily rises, while that of the egg-white increases until the 13th day. After the 13th day correlation between the changes of the material in the two fluids can no longer be pointed out. The *K content* increases in the amniotic fluid and decreases in the egg-white until the 18th day. At a 99% level of significance, until the 18th day the correlation of the material contents shows a value of 0.73. The tendency and significance of the temporal change are real, and not disturbed by the autocorrelation. The *acid phosphatase activity* of the egg-white suddenly increases on the 24th day, in connection with a higher rate destruction of the epithelia of the egg-white sac in the period of intensive egg-white swallowing, whereby the enzyme enclosed in the epithelia is released. The amylase activity keeps increasing until the 8th day, then suddenly drops, to remain at more or less the same level until hatching. No significant correlation between the substances of allantois and thick yolk was found.

The *albumen content* rises until hatching, both in the *allantoic fluid* and in the *egg-white*. Between the 18th and 21st day the correlation is significant. The correlation in time is not disturbed by any high rate autocorrelation. The increase of *lipid content* and decrease of *glucose content* shows a significant correlation until the 13th day. The quantitative change of lipid in time is significant, and not disturbed by autocorrelation. The *Na content* decreases until hatching in the allantoic fluid, and increases until the 18th day, then decreases in the egg-white. The change of correlation in time is significant until the 18th day, and not disturbed by autocorrelation.

The *Cl content* gradually decreases until hatching in the allantoic fluid, while increasing until the 13th day and decreasing afterwards in the egg-white. The correlation is significant until the 13th day. The *K content* decreases until the 18th day, then rises both in the allantoic fluid and in the egg-white. The correlation until the 12th day is significant at a 99% level of reliability, the correlation coefficient  $r = 0.71$ .

The total *protein content of the thick yolk* decreases until the 8th day, then increases until hatching; while that of the *thin yolk* increases from the 3rd to the 13th day, to decrease afterwards. The tendency of change in time is not, however, significant. The *albumen content* decreases in the thin and increases in the thick yolk between the 8th and 13th days, though at the end of incubation its concentration is lower in the latter. This correlation is significant (99%). The explanation lies in the absorption of the albumen content of the thin egg-white and the flow of material caused by the difference in concentration.

The *urea-N content* decreases in the thin and increases in the thick yolk. Changes in time show a significant correlation, and there is no autocorrelation. The *creatinine content* increases from the 8th to the 13th day in the thin yolk and decreases at the same time in the thick yolk. The tendency of change (path coefficient) is significant (90%). The *P content* decreases in the thick yolk until the growth of the thin yolk, then increases, while showing no quantitative change in the thin yolk. The tendency of change is significant. The *lipid content* decreases until hatching in the thick yolk and increases in the allantoic. At a 0.56 level of correlation, this change is verified by a 95 per cent reliability of correlation.

Increase in the K content of the thick yolk between the 8th and 13th days, with an unchanged amount of K in the thin yolk, is significant ( $R = 0.85$ ). In the thick yolk, the Cl content decreases from the 8th to the 13th day; while in the allantoic fluid, the amount of Cl steadily decreases until hatching. The correlation is valid at a 99% level of reliability. The path index on the 13th day is 0.73 at a 99% level of significance.

### Discussion

The flow of inorganic and organic matters in the embryo fluids has different ways due to the following morphological circumstances.

1. On the 3rd day of incubation the vitelline membrane ruptures; and then the flow of material directly between the yolk and the egg-white, and in the area vasculosa of the developing yolk sac, through its richly-veined wall by diffusion and active transport, is equally possible. Between the 3rd and 8th day of incubation, this flow is intensive and is accompanied by an increase in the volume of the so-called thin yolk. It is with this process that an increase in the Na, urea-N and glucose content of yolk and decrease in its K, Ca, lipid content and amylase activity are connected. In the meantime, the total protein content of the egg-white becomes greatly reduced while its glucose content and amylase activity increase.

2. The blood circulation of the yolk carries materials taken up from the yolk and egg-white to the embryo.



3. Besides an anastomosis between the blood circulation of the embryo and that of the yolk through the empty vein at the back, connection is established between the vascular system of the allantois on the one hand, and the blood vessels of the amnion and blood circulation of yolk sac at about its navel, on the other. Namely, at the edge of the navel, the blood vessels of the two vascular systems are in multiple anastomosis. The vascular system of the egg-white sac is, in fact, just one of those parts of the embryonic blood circulation which is close to the connection. In geese this anastomosis develops already on the 11th–12th day and is maintained until hatching. The anastomoses of the two blood vessel systems enable the yolk, the allantoic and amniotic fluids, as well as the substances taken up by the cells from the egg-white sac, to get equally into the same blood circulation. Thus, through the concentration differences and osmotic pressures of the contents of amniotic and allantoic fluids, yolk and egg-white, the common blood circulation makes the mutual flow of organic and inorganic matters contained in them indirectly possible, too.

4. In geese, the amniotic suture breaks on the 14th day, whereby the egg-white flows directly into the amniotic cavity. From the 17th to the 24th day, this flow is intensive, as indicated by the sudden increase in the protein and lipid contents of the amnion.

With all these taken into consideration, the significantly proved tendencies of flow of the organic and inorganic matters in the different fluids become morphologically acceptable, too. Water as a dissolving and conveying medium, together with the organic and inorganic matters dissolved in it, is the main carrier of material flow. Its flow between the 3rd and 8th day of incubation into the yolk sac, the yolk, then through the blood circulation of heart into the embryo, with the excretion of amniotic fluid into the amnion and by the functioning of the kidney into the allantois, is such an economical water circulation as is never produced in mammals and even in birds only during the period of hatching. The water requirements of material flow and embryonic growth until hatching are ensured by the embryonic blood circulation from the allantoic fluid. The water circulation of the bird developing in the egg is the most perfect and at the same time most vulnerable physiological process that must be primarily satisfied by the technology of incubation.

Quantitative changes during development in the organic and inorganic matters of embryonic fluids, as well as the development of embryo sacs, jointly prove that the prenatal development in geese is fractional.

The quantities of organic and inorganic matters of yolk, egg-white and embryonic fluids, and their changes in the course of development, are such basic data of hatching biology as can be made good use of in future diagnoses of technological faults of hatching.



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### References

- BAINTNER, K.—FEHÉR, GY. (1974): Fate of egg-white trypsin inhibitor and start of proteolysis in developing chick embryo and newly hatched chick. *Developmental Biology*, **36**, 272.
- EÖRY, A. (1976): VII. Neuman Kollokvium. Szeged.
- FÁNCSI, T.—FEHÉR, GY. (1979): Ultrastructural studies of chicken embryo chorioallantoic membrane during incubation. *Zbl. f. Vet. Med. Reihe C*, **3**, 370.
- FEHÉR, GY.—FÁNCSI, T.—MAJOROS, G. (1980): Ultrastructural studies of goose embryo chorioallantoic membrane (CAM). *Zbl. f. Vet. Med. Reihe C*, **9**, 363.
- FEHÉR, GY.—KÓTAI, I. (1980): A sziktoömlő ereinek és redőinek fejlődése és szerkezetváltozása házimadarokban (Development and structural change of folds and blood vessels of the yolk sac in domestic fowl). *Magy. Áo. Lapja*, **35**, 106.
- FEHÉR, GY.—TELKI, M.—FÁNCSI, T. (1980): A magzatburkok fejlődése, a magzati folyadékok, a fehérje és a szik mennyiségének változása a keltetés során lúdban (Development of the fetal membranes, quantitative changes of embryonic fluids, egg-white and yolk during incubation in geese). *Magy. Áo. Lapja*, **35**, 761.
- ILJIN, M. D. (1917): Materials for embryo chemistry. Archiv. Leningrad.
- JACKEZ, J. A. (1972): Compartmental analysis in biology and medicine. Elsevier Publ. Comp., New York.
- NEEDHAM, J. (1963): Chemical embryology I—III. New York—London. Hafner Publ. Comp.
- RAPPAPORT, L.—EICKHORN, M. (1960): Medizinische Biochemie VEB.
- ROMANOFF, A. L. (1960): The avian embryo. The Mcmillan Company, New York.
- SCHALES, O. (1941): Methods satellites in biology. *Journ. Biol. Chem.*, **140**, 875.
- SÓS, J. (1974): Laboratóriumi diagnosztika (Laboratory diagnostics). Medicina, Budapest.
- SZÉKELY, L.—BARTALITS, L. (1978): Satellite módszer gyűjtemény (Satellite method collection). Medicina Kiadó, Budapest.
- WENDEL, T. (1955): Compartmental analysis in the amnion and allantois fluids. *Amer. Journ. Chick Pathol.*, **25**, 840.
- ZÖLLNER, N.—KIRSCH, K. (1962): Einige biochemische Methoden für Embryologie. *Zeitschr. Gen. exp. med.*, **135**, 545.



## RELATIONSHIP BETWEEN WATER SUPPLY AND ASSIMILATION IN APPLE TREES

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The authors studied the photosynthesis and transpiration as a function of soil moisture in Starkrimson Delicious apple trees on M9 rootstock. The relationship between daily net photosynthesis ( $P_n$ ) and soil moisture can be expressed by the quadratic equation  $y = a + bx - cx^2$ . The values of the regression coefficient are 0.76 for the relationship between  $P_n$  and soil moisture, and 0.89 for that between the assimilation balance for 24 hours and soil moisture. Net photosynthesis before noon is not so strictly regulated by the soil moisture as  $P_n$  for the whole day. As the soil water content decreases  $P_n$  also decreases and dark respiration increases. At higher soil moisture the increase in the assimilation balance is higher than that in transpiration. Dry matter production and utilization of soil moisture by the apple variety examined is better when the soil contains more water.

### Introduction

Hungarian and international literature on the physiology of fruit trees provides relatively few data on how the life processes, particularly  $\text{CO}_2$  assimilation and transpiration, of the tree are influenced by the water content of the soil. One of the most important questions when developing irrigation techniques is when and to what extent the water requirements of the plants should be satisfied by irrigation under given conditions, if the operation is to be economical and production aims are taken into consideration (CSELŐTEI 1965). It is necessary to determine the amount of water required to obtain the average yields of various plant species, and the yield level at which a natural water supply restricts the normal life processes of the plants. In Hungary the relationship between water consumption, ecological factors and dry matter production have been comprehensively analysed by CSELŐTEI (1965) for vegetables and MIHÁLYFALVY (1970) and SZALÓKI (1968, 1970) for field crops.

For perennial and arboraceous plants the relationship between dry matter production and transpiration is more difficult to determine than in the case of annual crops. Nor is it always possible to employ research methods suitable for herbaceous plants without modification when studying trees. This is why only a few researchers have undertaken to clarify the basic relationship between water consumption and dry matter production in fruit-trees. CERVENKA (1970) obtained maximum assimilation in apple trees with a uniform, satisfactory



water supply. As the soil water content decreased, the water deficiency of the leaves increased, while transpiration and assimilation gradually decreased. According to PETINOV (1954) the correct values of the lower and upper limits of optimum water supply are exhibited by the physiological responses of the plant itself. In order to discover the most important correlations between water regime and photosynthesis in plants the radiation-energetic balance of the orchards must be investigated (NICHIPOROVICH—CHMORA 1963). In experiments by KUSHNIRENKO (1975) the intensity of photosynthesis in the apple variety Papirovka was sharply reduced by drought. This author considers the water supply to be one of the most important factors acting on the status of the photosynthetic pigment system. There was a considerable difference in the intensity of transpiration between apple trees raised at 70% and 30% soil water capacity (110 and 86 g/m<sup>2</sup>/hour); if the dry matter accumulation is taken into consideration, trees with a better water supply and increased transpiration showed higher dry matter production (4.99 and 3.23 g dry matter/1000 g water respectively for the treatments mentioned above). SLOWIK (1974) found three possibilities concerning the water uptake of fruit trees:

1. the available water content of the soil (AW) between full water capacity (FW) and dead water content (DW) can be taken up without any difficulty;

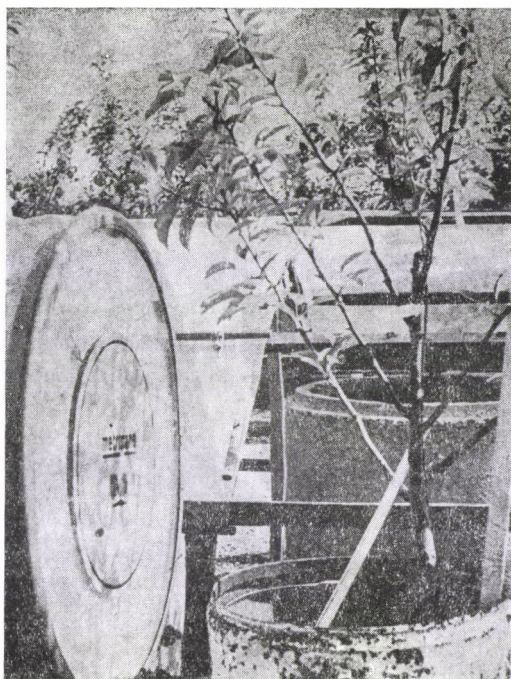
2. AW can be taken up easily to a certain value, then the rate of water uptake decreases;

3. with a gradual decrease in the water content, water uptake becomes more and more difficult. The aim of the present paper was to examine these little known correlations by measuring the assimilation and transpiration of apple trees under controlled soil moisture conditions.

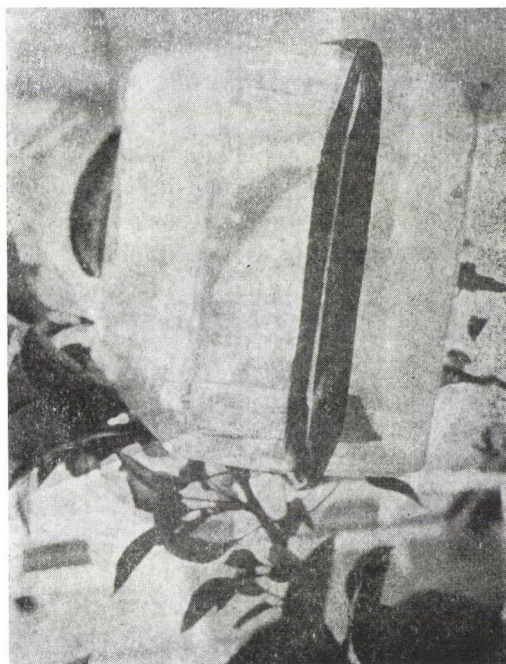
### Material and method

Two-year-old Starkrimson Delicious apple trees on M9 rootstock, raised in pots with a 50 kg dry soil capacity were used in the experiment. The roots completely enmeshed the soil in the pots and the trees had a leaf area of about 0.2 m<sup>2</sup>. The soil used in the experiment was a prairie loam with a water capacity of 28.6% by weight and a dead water content of 11.8% by weight. The pots were saturated with water to full capacity and covered with plastic film to prevent evaporation. The transpiration of the trees could thus be determined from the reduction in weight registered every 24 hours. The measurement of assimilation was started at various dates from the end of July to mid-August, so the activity of trees with different soil moisture levels was measured as a function of the drying process in pots saturated to water capacity at various dates. When the dead water content was reached and water uptake ceased, the dead water content, the dry weight of the soil, and the weights of the pot, the plant, etc. were determined, so that the water content of the soil could be calculated at any time during the experimental period.

The values of assimilation and dark respiration were determined by means of an Infralyt-4 (GDR, Junkalor) infrared gas analyser. The 24-hour dry matter production balance was calculated from the difference between net photosynthesis in day-time and dark respiration. The leaves were placed in transparent plexy vessels with a capacity of 2.6 l, and an air flow of 36 l/hr. Dust-free air dried with silica gel was conducted into the measuring instru-



*Fig. 1. One of the pot-grown experimental trees in the course of weighing*



*Fig. 2. The plexiglass vessel and the leaves examined*



ment, which registered the difference in  $\text{CO}_2$  content between the surrounding (control) air and that flowing through the leaf space (measuring site) in ppm. Data on air humidity and radiation were obtained from the Agrometeorological Observatory situated 3 km from the experimental site. Soil moisture data were obtained by exsiccator tests at 105 °C. Figure 1 shows one of the pot-grown experimental trees in the course of weighing. In Fig. 2 the vessel used in the net photosynthesis examination and the leaves examined are seen.

## Results

The carbon dioxide turnover in the leaves of apple trees on a meteorologically characteristic day showed the trend seen in Fig. 3. The continuous line shows the net photosynthesis and respiration of a tree well supplied with water, while the broken line represents a tree with a reduced water supply.

The figure shows that the net photosynthesis can be depicted by a two-peak curve, and that in a tree well supplied with water the photosynthetic activity is higher and the respiration lower than in a tree with a reduced water supply.

Net photosynthesis as a function of the moisture content of the soil is shown in Fig. 4.

The relationship between soil moisture and net photosynthesis can be best described by the quadratic equation  $y = a + bx - cx^2$ , which indicates that with an increase in the soil water content the photosynthetic activity increased steadily, though to a lesser extent.

The correlation seen in Fig. 5 shows that the net photosynthesis in the period preceding the depression at noon (from 5 to 11 a.m.) is less sensitive to changes in the soil water content.

Of the meteorological factors global radiation exercised the greatest effect on the total daily net photosynthesis. The relationship is shown in Fig. 6.

According to the correlation analysis the relation is positive and linear in the range of radiation examined (250–500 cal/cm<sup>2</sup>/day).

The relationship between the intensity of transpiration as a dependent variable, and soil moisture and air temperature as independent variables is shown by the three-dimensional graph in Fig. 7.

The correlation coefficient  $r_{x_{3y}}$  indicates the closeness of the binary relationship between transpiration and global radiation. From this it can be concluded that with a rise in the water content of the soil the intensity of transpiration continues to increase, though to a lesser extent.

The total daily net  $\text{CO}_2$  assimilation, i.e. the dry matter accumulation, and the intensity of transpiration as a function of the water content of the soil are seen in Fig. 8.

The figure shows that the relationship between net  $\text{CO}_2$  assimilation and soil moisture can also be described by a quadratic equation, i.e. it shows saturation character. Comparing the 24-hour assimilation balance with the



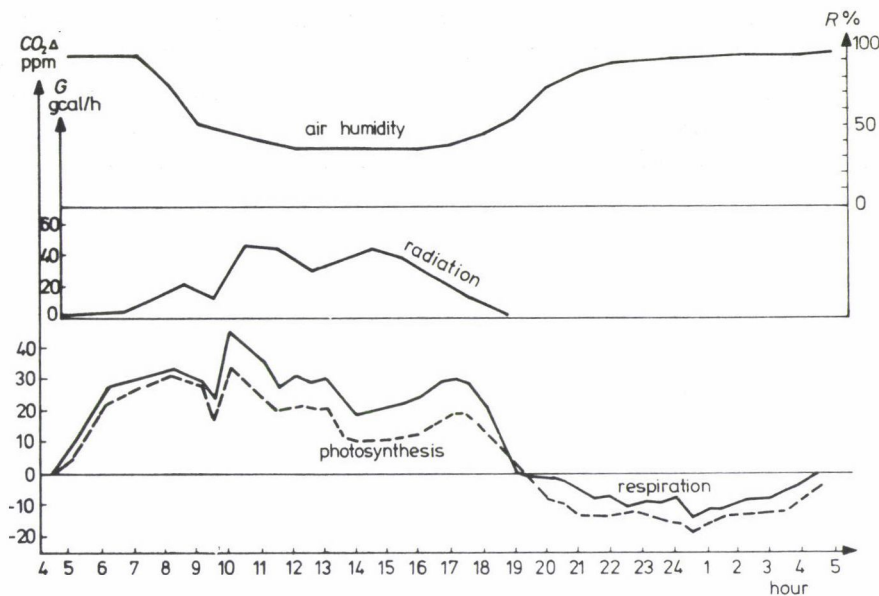


Fig. 3. Net photosynthesis and dark respiration of Starkrimson Delicious apple trees on M9 rootstock at different water supply levels, with radiation and air humidity indicated. Erd-Elvira Research Station, 26th August 1977

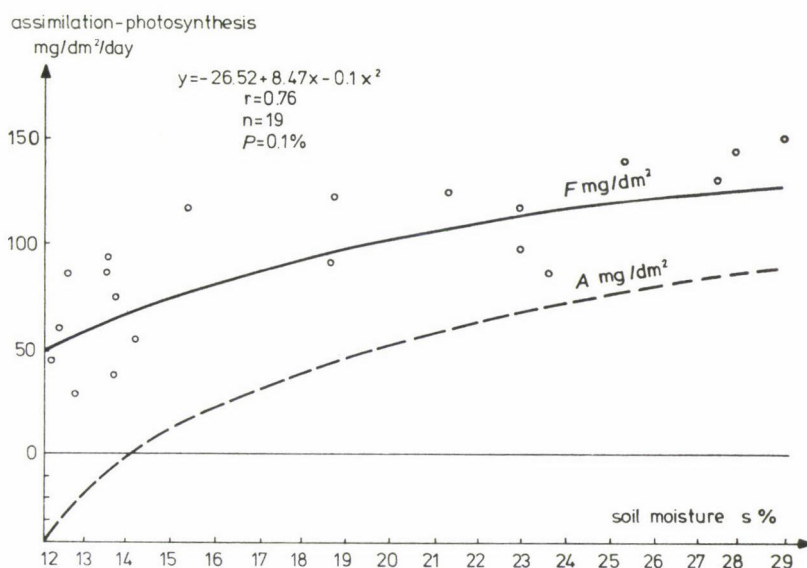


Fig. 4. Net photosynthesis in leaves of Starkrimson Delicious apple trees on M9 rootstock ( $F$ ;  $\text{mg CO}_2/\text{dm}^2$  leaf/day) as a function of soil moisture

character of the change in the transpiration intensity it can be established that at higher levels of soil moisture the intensity of transpiration increases to a lesser extent relative to the increase in assimilation. Thus, parallel with an increase in the moisture content within the water capacity of the soil the accumulation of assimilates per unit transpiration shows a continual increase.

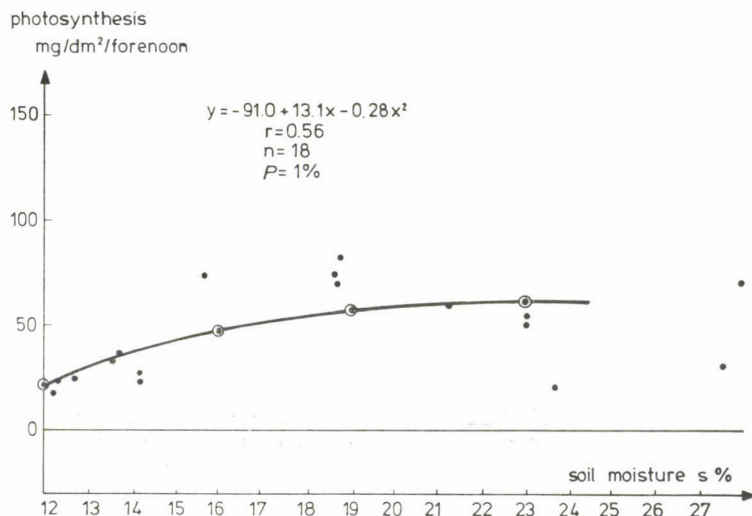


Fig. 5. Total net photosynthesis of Starkrimson Delicious apple trees before noon as a function of soil water content

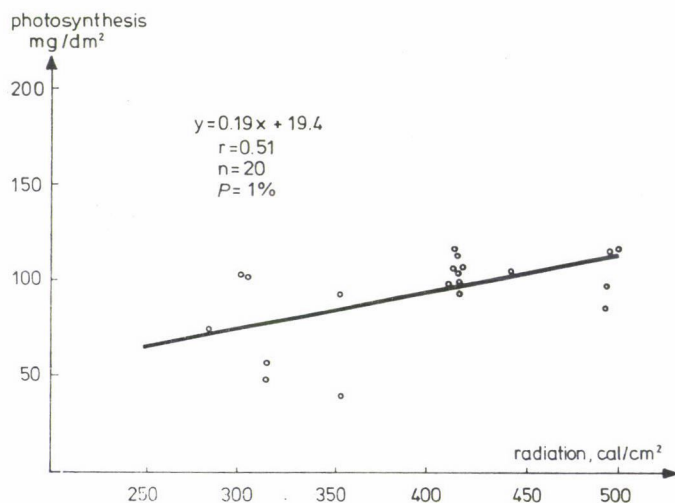


Fig. 6. Total daily net photosynthesis of Starkrimson Delicious apple trees as a function of radiation

A favourable water supply involves higher water consumption, but at the same time results in better water utilization due to the increased dry matter production. This is also indicated by the lower transpiration coefficient. The calculated values of the regression curve for the relation between assimilation and soil moisture are represented by the continuous line (1), while the broken

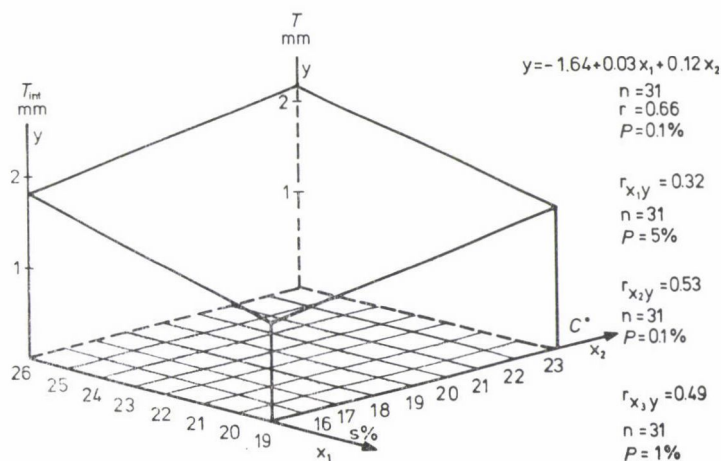


Fig. 7. Transpiration of leaves of Starkrimson Delicious apple trees ( $\text{l/m}$ ) as a function of soil moisture (% by weight), air temperature ( $^{\circ}\text{C}$ ) and global radiation ( $\text{cal/cm}^2/\text{hour}$ )

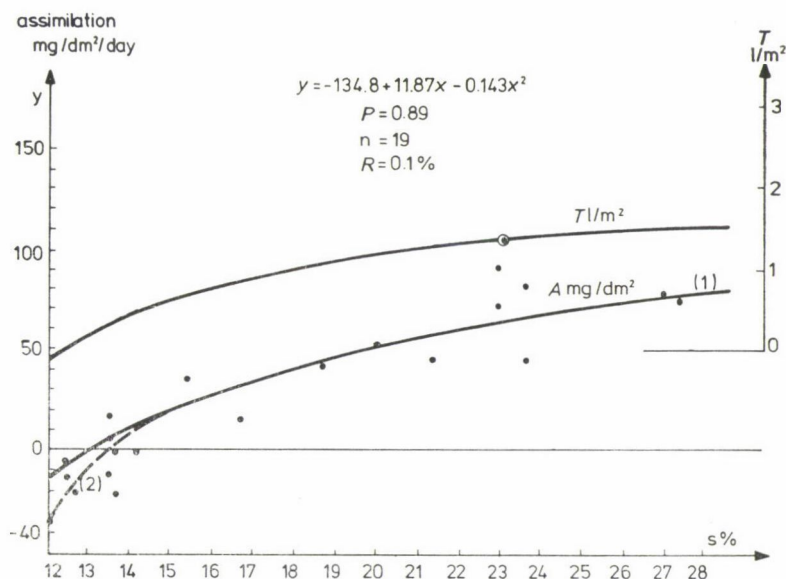


Fig. 8. 24-hour balance of assimilation ( $A$ ), and transpiration ( $T$ ) as a function of soil moisture in Starkrimson Delicious apple trees



line (2) shows the trend of the measured values. The point of compensation, where the dry matter production and utilization are equal, is found at a soil moisture value of 13.3%, which corresponds to about 10% available water content.

### References

- CERVENKA, K. (1970): Zmeny obsahu vody a asimilace  $\text{CO}_2$  v listach podnoz M9 pri klesajúcim obsahu vody v pude. *Zahradnictvi*, **1**, 17–20.
- CSELŐTEI, L. (1965): Az öntözés rendszerének tényezői a zöldségnövényeknél (Factors of the irrigation system in vegetable crops). D.Sc. Thesis, Manuscript. 1–332.
- KUSHNIRENKO, M. D. (1975): Fiziologija vodobmena i zasuhoustojkivosti plodovüh rastenij. Kisinyev, 1–216.
- MIHÁLYFALVY, I. (1970): Fő- és másodvetésű növények vízfogyasztásának, vízhasznosításának vizsgálata (Water consumption and utilization in main and second crops). *Öntözéses Gazdálkodás*, **8**, 1, 3–15.
- NICHIPOROVICH, A. A.—CHMORA, S. S. (1963): Vodnűj režhim v svazi s z obmenom veshestv i produktivnosztju. Moscow, 1–114.
- PETINOV, N. S. (1954): Fiziologija rastenij. Moscow, 1–179.
- SŁOWIK, K. (1974): Water relations in soils, salinity, pH. *Proc. XIX. Int. Hort. Congr.* Warsaw, III. 335–342.
- SZALÓKI, S. (1968): A talajvíz-mélység hatása a lucerna termésalakulására és vízgazdálkodására (Effect of groundwater level on yield trend and water regime in alfalfa). *Öntözéses Gazdálkodás*, **4**, 57–75.
- SZALÓKI, S. (1970): A cukorrépa levélzetének, gyökértermésének és vízfogyasztásának kapcsolata (Relationship between foliage, root yield and water consumption in sugar-beet). *Öntözéses Gazdálkodás*, **8**, 61–74.

## EFFECT OF PRE-SOWING "VITAMINE B" SEED TREATMENT ON EATING PAPRIKA

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### Introduction

Studies on the stimulative effect of vitamins on the development of plants began in the second half of the thirties (KÖGL—HAAGEN—SMITH 1936). Growth stimulation by vitamins was first studied with isolated organs (root, embryo) (ROBBINS—BARTLEY 1937, WENT—THIMANN 1937, BONNER 1938, ADDICOT—DEVIRIAN 1939, BONNER 1940). Then, from 1950, the whole plant was subjected to examination with the different phases of development taken into consideration.

The effect of vitamins belonging to group "B" on the yield, and value of plant components, was initially investigated by Soviet authors. OVCSAROV (1969) gives a full literary summary of the work related with vitamins. Accordingly, the yield- and composition responses of plants to treatments with vitamins of group "B" were first studied in 1960-1964 for maize (SINKOVICS 1963, 1964a, b, 1965a, b, 1967, STOLETOV *et al.* 1963), in 1968-1970 for sweet melon (SINKOVICS 1970a, b), and in 1968-1971 for eating and spice paprika (SINKOVICS 1972, 1974).

As known from literary data, the vitamins of group "B", in their biological effect, are biocatalytic substances possessing the highest activity. Their role is mainly known in the activity of enzymes, where they have definite functions in developing the different properties.

The vitamins are formed through biosynthesis, mostly in the leaves during the development of plants, and always move to organs performing the most important life processes, where they accumulate in relatively large quantities (STOLETOV *et al.* 1963, SINKOVICS 1967).

Only under optimum conditions are plants able to synthesize vitamins, but this amount is probably insufficient for the development of the economically important characteristics. For this reason, we intend to try improving the early maturity and yield of eating paprika, one of the most important vegetable plants in Hungary, by treating the seed with vitamin solutions.

### Material and method

The experiments were carried out with the eating paprika varieties Javított Cecei and Csokros Konzerv on the Budatétény ground of the Horticultural Research Institute in 1967–1971. For the treatments, various concentrations and combinations of meso-inosite, nicotinic acid, cobalamine, biotine, thiamine, pyridoxine and panthetonic acid were used (Table 1).

The seed treatments were carried out with the technique elaborated by us and licensed by the National Planning Bureau in 1974. The experiments were laid out in a Latin block

**Table 1**  
*Vitamins used in seed treatments*

Vitamins	Concentration, %		
	1.0	0.1	0.05
Meso-inosite	1.0	0.1	0.05
Nicotinic acid	0.1	0.01	—
Cobalamine	0.001	0.0001	—
Biotin	0.01	0.001	—
Thiamine	0.1	—	—
Pyridoxine	0.1	—	—
Panthotenic acid	0.1	—	—

design in 4 replications, with 56 plants per plot shown to a  $50 \times 20 + 20$  cm alternating twin-row pattern. On setting out the seedlings, we recorded the plant heights, the number of leaves, and the fresh and dry weights of leaves (for 100 plants per treatment). At the time of flowering, we recorded the number of flowered on 3 occasions with 10-day intervals.

Early maturity was determined on the basis of surplus yield compared with the untreated control of the first picking. The fruit was sorted in grades I, II and III, and waste. In the tables the weights of fruit from the first picking, and the total fruit weight (of the fourth to fifth picking) are given in t/ha and %, and the % of the I grade produce in comparison with the control, then the yield difference in t/ha. The figures show the development of seedlings and fruit. The data of the experiments were evaluated by analysis of variance.

### Results

Vitamins belonging to group "B", when applied at a proper concentration in the form of seed treatment, significantly increased the early maturity and yield of the eating paprika. The effect of seed treatments could already be seen at the time of raising the seedlings. Treated seeds germinated 2–3 days earlier than the untreated ones, and the seedlings obtained from them were, when set out stronger and more viable, and had well developed roots (Fig. 1).

The dry weight of seedlings in the meso-inosite 0.1 and nicotinic acid 0.01 treatments increased by 76–60% in the variety Javított Cecei and by 51–41% in Csokros Konzerv, compared with the control (Figs 2 and 3).

This means that the initial growth of seedlings was better and their development faster, followed by the intensive development of reproductive organs. In the meso-inosite 0.1 and nicotinic acid 0.01 treatments, the number



of flowers per plant of the variety Javított Cecei in the first period of flowering was 90–95% higher than in the control (Fig. 4).

In 1969 the yield average of the first picking in the variety Javított Cecei rose significantly, by 63% (4.46 t/ha) in the meso-inositol 0.1 treatment and by 37% (2.62 t/ha) in the nicotinic acid 0.01 treatment. In these treatments the weight of the grade I berries was 140–59% higher than in the control (Table 2). Seed treatment with a vitamin solution of nicotinic acid 0.01 +

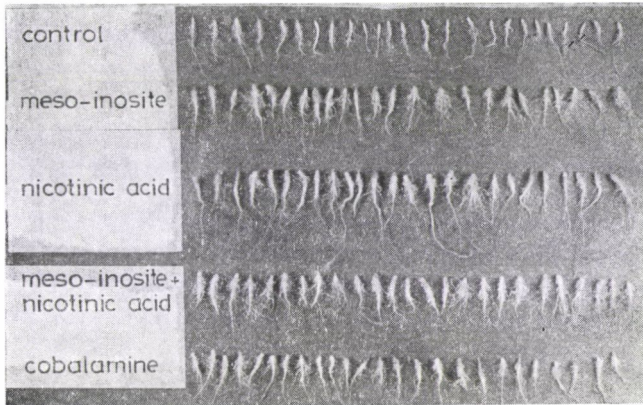


Fig. 1. Roots of seedlings of the paprika variety Javított Cecei in seed treatments with vitamin solutions, 1970

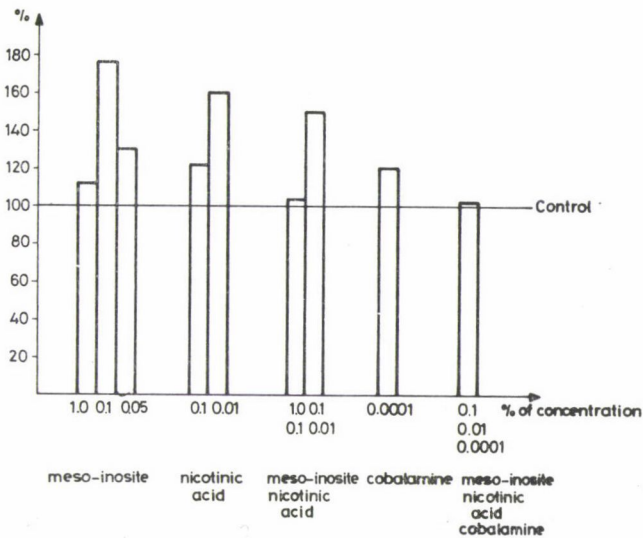


Fig. 2. Dry weight of seedlings of the paprika variety Javított Cecei when set out, as percentage to the control, 1970

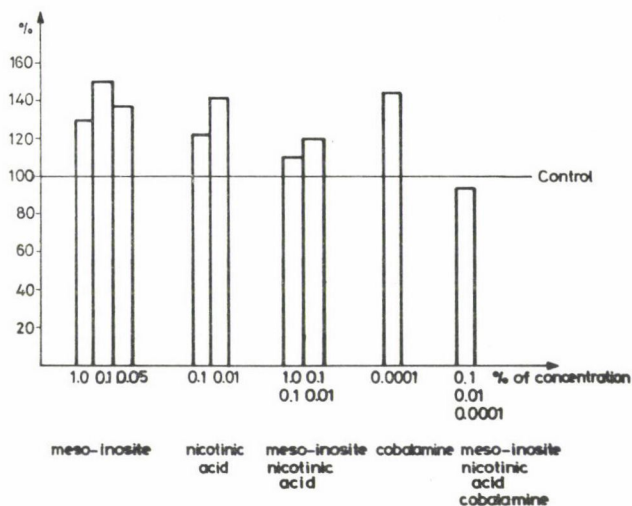


Fig. 3. Dry weight of seedlings of the paprika variety Csokros Konzerv when set out, as percentage to the control, 1970

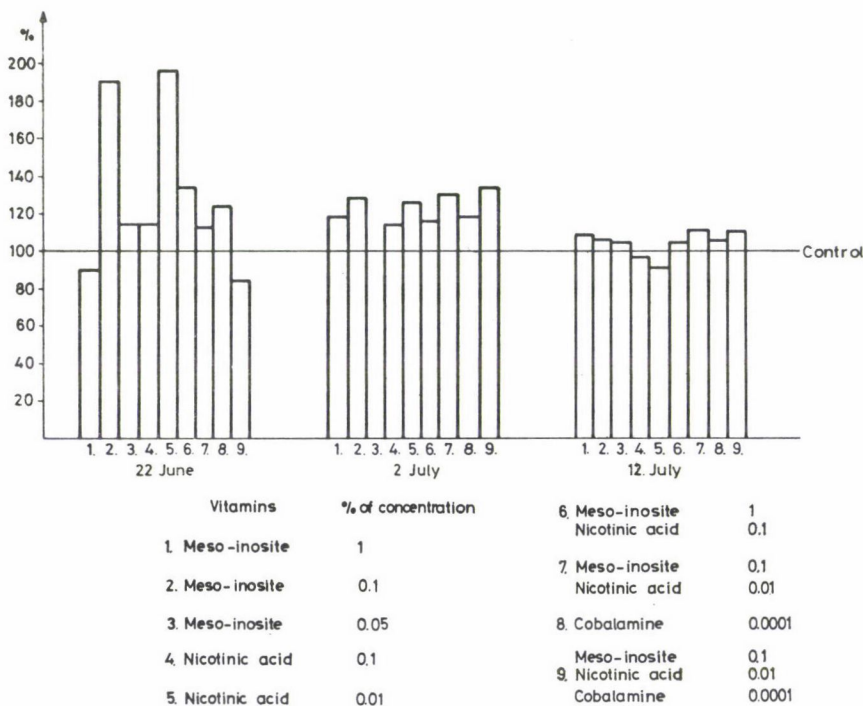


Fig. 4. Result of observation of the paprika variety Javitott Cecei in 1970

**Table 2**  
*Effect of seed treatment with vitamin solution on early maturity  
 and yield in the eating paprika variety Javított Cecei  
 (1969)*

Treatments	Concentra- tion, %	First picking 11th August				Picking total			
		Yield		I.-grade berry, %	Yield difference, t/ha	Yield		I.-grade berry, %	Yield difference, t/ha
		t/ha	%			t/ha	%		
Control		7.04	100	100	—	19.96	100	100	
Meso-inosite	0.1	11.50	163	240	+4.46***	24.87	125	190	+4.91*
Nicotinic acid	0.01	9.66	137	159	+2.62*	21.75	109	144	+1.79
Pyridoxine	0.1	6.91	98	85	-0.13	20.38	102	100	+0.42
Cobalamine	0.0001	7.66	109	89	+0.62	21.21	106	99	+1.25
Thiamine	0.1	8.88	126	90	+1.84	23.33	117	104	+3.37
Nicotinic acid	0.01								
Thiamine	0.1	5.18	74	65	-1.86	20.93	105	99	+0.97
Biotin	0.001								
* SD <sub>5%</sub>					2.29	4.69			
** SD <sub>1%</sub>					3.06	—			
*** SD <sub>0.1%</sub>					4.2				

**Table 3**  
*Effect of seed treatments with vitamin solutions on early maturity  
 and yield in the eating paprika variety Javított Cecei  
 (1970)*

Treatments	Concentra- tion, %	First picking 3rd August				Picking total			
		Yield		I.-grade berry, %	Yield difference, t/ha	Yield		I.-grade berry, %	Yield difference, t/ha
		t/ha	%			t/ha	%		
Control		4.03	100	100	—	29.48	100	100	—
Meso-inosite	1.0	3.82	95	119	-0.21	28.22	95	127	-1.26
Meso-inosite	0.1	6.29	156	241	+2.26**	32.50	111	172	+3.02
Meso-inosite	0.05	4.10	102	146	+0.07	28.35	96	146	-1.13
Nicotinic acid	0.1	3.99	97	130	-0.04	26.96	91	141	-2.52
Nicotinic acid	0.01	6.43	160	189	+2.40***	30.85	105	147	+1.37
Cobalamine	0.0001	4.05	101	132	+0.02	28.66	97	136	-0.82
Meso-inosite	1.0	4.47	104	154	-0.14	30.68	104	174	+1.20
Nicotinic acid	0.1								
Meso-inosite	0.1	5.12	127	181	+1.09	28.87	98	170	-0.61
Nicotinic acid	0.01								
Meso-inosite	0.1								
Nicotinic acid	0.001	2.50	62	67	-1.53*	25.76	87	131	-3.72
Cobalamine	0.0001								
* SD <sub>5%</sub>					1.35	5.23			
** SD <sub>1%</sub>					1.84	Yield difference in picking total is not significant			
*** SD <sub>0.1%</sub>					2.4				



thiamine 0.1 + biotin 0.01 had a negative effect on early maturity. As for the total yield, only the meso-inositol 0.1 treatment showed significant difference compared with the control.

In 1970 the yield of the first picking in the variety Javított Cecei was significantly larger in the meso-inositol 0.1 and nicotinic acid 0.01 treatments by 56 and 60% (2.26 and 2.4 t/ha). The weight of berries in grade I was 141–80% higher than in the control (Table 3). Difference in total yield was not significant. The application of a combined solution of meso-inositol and nicotinic acid did not cause any significant increase in yield. Treatment with a solution of meso-inositol + nicotinic acid + cobalamine had a negative effect on yield, as regards both the first and the total crop.

In the variety Csokros Konzerv, the yield of the first picking in 1970 in the meso-inositol 0.1 treatment was significantly (38%, 1.69 t/ha) higher than in the control (Table 4). The photos confirm the success of treatments (Fig. 5). The weight of berries in grade I exceeded that of the control by 75%. The 1.0–0.05% solution of meso-inositol had a positive effect in this variety, but the difference in yield was insignificant. In the nicotinic acid 0.1 and 0.01

Table 4

*Effect of seed treatments with vitamin solutions on early maturity and yield in the eating paprika variety Csokros Konzerv (1970)*

Treatments	Concentration, %	First picking 3rd August				Picking total			
		Yield		I.-grade berry, %	Yield difference, t/ha	Yield		I.-grade berry, %	Yield difference, t/ha
		t/ha	%			t/ha	%		
Control		4.39	100	100	—	12.00	100	100	—
Meso-inositol	1.0	5.05	115	162	+0.66	13.03	109	159	+1.33
Meso-inositol	0.1	6.08	138	175	+1.69**	13.18	110	159	+1.16
Meso-inositol	0.05	5.58	127	173	+1.19	13.15	109	141	+1.15
Nicotinic acid	0.1	5.40	123	174	+1.01	12.45	104	176	+0.45
Nicotinic acid	0.01	5.05	115	172	+0.66	11.78	98	134	−0.22
Meso-inositol	1.0	3.69	84	145	−0.70	11.97	99	159	−0.03
Nicotinic acid	0.1								
Meso-inositol	0.1	4.19	81	175	−0.86	12.56	105	134	+0.56
Nicotinic acid	0.01								
Cobalamine	0.0001	5.73	130	185	+1.34	15.15	126	143	+3.15**
Meso-inositol	0.1								
Nicotinic acid	0.01	3.39	77	67	−1.00	13.43	112	164	+1.43
Cobalamine	0.0001								

\* SD<sub>5%</sub>

\*\* SD<sub>1%</sub>

1.52

—

1.81

2.41

treatments, the 23 and 15% yield increase on the first picking was insignificant. Cobalamine at a concentration of 0.0001% exercised a positive effect on early maturity. On first picking, the yield was higher by 30% (1.34 t/ha) than in the control. As for the total yield a significant (26%, more than 3 t/ha) difference could be observed.

In 1971 it was again with the meso-inosite 0.1 treatment that the variety Csokros Konzerv showed the best result. The increase in yield was 77% (2.04 t/ha) on first picking, and the grade I produce exceeded the control by 93%.



Fig. 5. Plots of the eating paprika variety Csokros Konzerv before the first picking in 1970

Table 5

*Effect of seed treatments with vitamin solutions on early maturity and yield in the eating paprika variety Csokros Konzerv (1971)*

Treatments	Concentration, %	First picking 20th July				Picking total			
		Yield		I.-grade berry, %	Yield difference, t/ha	Yield		I.-grade berry, %	Yield difference, t/ha
		t/ha	%			t/ha	%		
Control		2.69	100	100	—	14.09	100	100	—
Meso-inositol	1.0	3.87	144	249	+1.18	14.02	100	121	-0.07
Meso-inositol	0.1	4.78	177	193	+2.09**	15.58	110	112	+1.49
Nicotinic acid	0.1	3.93	145	181	+1.24	13.67	97	106	-0.42
Biotin	0.01	2.85	108	146	+0.16	13.93	99	108	-0.16
Biotin	0.001	2.97	109	150	+0.28	14.47	103	121	+0.38
Pyridoxine	0.1	4.27	158	152	+1.58*	14.82	105	119	+0.73
Phanthotenic acid	0.1	3.35	124	235	+0.66	15.37	109	114	+1.28
Cobalamine	0.0001	4.06	151	225	+1.37	16.73	118	135	+2.64*
Thiamine	0.1	3.40	126	158	+0.71	14.66	104	128	+0.57
* SD <sub>5%</sub>					1.48	2.17			
** SD <sub>1%</sub>					1.95	—			

Cobalamine at a concentration of 0.0001 — as in the previous year — increased the volume of yield both on first picking and on the average of all pickings (1.37 and 2.64 t/ha, respectively). The effect of 0.1% pyridoxine on early maturity was also remarkable. On first picking, the berry weight increased significantly — by 58% (1.58 t/ha); grade I produces were 52% more than in the control (Table 5).

### Discussion

Among the positive effects of seed treatments with vitamin solutions, *early maturity* is the most decisive one. In terms of time this means that the first yield can be picked 5–6 days earlier from the treated plants than from the control. The high number of flowers recorded in the first phase of flowering is in linear correlation with the volume of yield on first picking.

The effect of vitamins on early maturity is manifest in the significantly larger yield on first picking, but the surplus of the total yield is not always significant. On the basis of several years of experiments we have established that, if on first picking the number of berries is 40% higher than in the control, a decrease in yield should be reckoned with on the second picking. This is probably due to the fact that, for the development and growth of the first berries, large amounts of nutrients are required: consequently, the subsequent



flowers cannot sufficiently develop. This yield reduction could probably be counterbalanced by an increased nutrient supply, as shown in an experiment with sweet melon (SINKOVICS 1970a, b).

Some vitamins may exercise either a positive or a negative effect on early maturity and yield, depending on the concentration. For example, in the case of the variety Javított Cecei, meso-inositol has the most positive effect at a concentration of 0.1%; with an increase or decrease in the concentration, the effect lessens, or even becomes negative on the total yield (Table 3).

As for the variety Csokros Konzerv, meso-inositol — while increasing the early maturity and yield at both higher and lower concentrations — is the most effective at the 0.1% concentration (Table 4).

When using vitamins, in addition to the concentration, the vitamin demand of the variety in question must also be considered. For example, pyridoxine had a negative effect on early maturity in the variety Javított Cecei, while significantly increasing this quality of the variety Csokros Konzerv. The vitamins have a negative effect on plants when the functional groups are transformed into isosteres (e.g. the OH group into a CH or NH group) and antivitamins are formed. The antivitamins become toxic when combined with the same proteins as the vitamins have. This causes more or less disturbance to the metabolic processes, and may ultimately lead to destruction of the plants.

Besides the early maturity the size of berry is worth mentioning. In the most efficient treatments the weight of fruit in grade I was nearly double of that of the control. Increase in yield is, however, determined first of all by the number of berries; the size of berry is but a secondary factor.

As to the positive effect of vitamins, it can be supposed that the vitamins as coenzymes influence the metabolism in a positive direction. Thus, the development of the whole plant, the formation, growth and development of the reproductive organs is hastened; and as a consequence, the first berries of significantly increased number, weight and size can be picked earlier.

## References

- ADDICOT, F. T.—DEVIRIAN, P. S. (1939): A second growth factor for excised pea roots; nicotinic acid. *Amer. J. Bot.*, **26**, 667–672.
- BONNER, J. (1938): Nicotinic acid and the growth of isolated pea embryos. *Plants physiology*, **13**, 865–869.
- BONNER, J. (1940): On the growth factor requirements of isolated roots. *Amer. J. Bot.*, **27**, 692–697.
- KÖGL, F.—HAAGEN-SMITH, A. J. (1936): Biotin und Aneurin als Phytoplants. *Zeitschr. f. physiol. Chem.*, **243**, 209–214.
- OVCSAVOR, K. J. (1969): Vitaminü rasteinij. *Izd. Kolos. Moscow.*
- ROBBINS, W. J.—BARTLEY, M. A. (1937): Vitamin B<sub>1</sub> and the growth of excised tomato roots. *Science*, **85**, 246–252.

- SINKOVICS, M. (1963): "B" csoportba tartozó vitaminok vizsgálata hibridkukoricákban (Study on vitamins of group "B" in maize hybrids). "Kukorica nemesítési és termesztési szimpozium". MTA Mezőgazdasági Kutatóintézete, Martonvásár.
- SINKOVICS, M. (1964a): Vlijanie vitaminov gruppü "B" na process oplodotvorenija kukuruzü. Kand. Diss. M.G.U. Moscow.
- SINKOVICS, M. (1964b): Nakoplenie vitaminov gruppü "B" razvivajuschih cemenah kukuruzü. Kukuruz, 6, 43-45.
- SINKOVICS, M. (1965a): A "B" csoportba tartozó vitaminok szerepe a heterózis jelenségében (Role of vitamins belonging to group "B" in the phenomenon of heterosis). Növénytermelés, 16, 173-179.
- SINKOVICS, M. (1965b): Nakoplenie vitaminov gruppü "B" v gibride kukuruzü i v roditelszkih formah. Naucsñie dokladü vüzsej skolü. Biologiceszkie Nauki., 1, 186-188.
- SINKOVICS, M. (1967): "B" csoportba tartozó vitaminok felhalmozódásának vizsgálata a kukorica növény vegetatív és generatív szerveiben (Accumulation of vitamins of group "B" in the vegetative and generative organs of the maize plant). Növénytermelés, 16, 247-256.
- SINKOVICS, M. (1970a): "B" csoportba tartozó vitaminok hatékonyságának növelésével kapcsolatos vizsgálatok sárgadinnyénél (Investigations to increase the efficiency of vitamins of group "B" in sweet melon). Zöldségtermesztés, 4, 125-134.
- SINKOVICS, M. (1970b): "B" csoportba tartozó vitaminok hatása a sárgadinnye termős virágai kialakulására és termőképességére (Effect of vitamins belonging to group "B" on the development and fertility of female flowers in sweet melon). Zöldségtermesztés, 4, 157-164.
- SINKOVICS, M. (1972): A "B" csoportba tartozó vitaminok hatásának vizsgálata néhány kertészeti növénynél (Effect of vitamins belonging to group "B" on some horticultural plants). A Magyar Biológiai Társaság X. Vándorgyűlésének programja, Szeged.
- SINKOVICS, M. (1974): A method of treatment with vitamin solutions for eating paprika. Acta Agron. Hung., 23, 410-413.
- SINKOVICS, M. (1974): Eljárás paprika és sárgadinnye vetés előtti magkezelésére. Szabadalmi leírás. Száma: 162269. Nemzetközi osztályozás: A 01 C 1/00. (A technique of pre-sowing seed treatment of paprika and sweet melon. Patent description. No. 162269. International grading: A 01 C 1/00.)
- STOLETOV, W. N.—ODINCOVA, JE. N.—SHINKOVICH, M. (1963): The phenomenon of heterosis and vitamin concentration in maize plant. Genetics Today. Proceedings of the XI. International Congress of Genetics, The Hague, The Netherlands, I.

## VARIA

### FRUIT SET IN APRICOT VARIETIES

Apricot production involves considerable problems caused by the low ecological tolerance and high agrotechnical requirements of the varieties currently used in commercial production. Further difficulties arise from the fact that the producers are not sufficiently acquainted with the biology of fruit set in the different varieties.

The experiments were therefore aimed at studying the characteristics of fruit set in apricot varieties in order to acquire knowledge of the nature of

- self- and cross pollination,
- geitonogamy, and
- spontaneous pollination (of free-standing flowers).

The investigations were aimed at demonstrating the necessity of mixing varieties, and at giving an answer to the question of which varieties are self-sterile or "partially self-fertile" and which are the most suitable pollen donors.

Very few authors in Hungary have dealt with the problems of fruit setting in apricot trees.

According to HORN (1939)-most apricot varieties are self-fertile, though no data were published to support this statement.

MALIGA (1966) found the variety Ananas to be self-sterile, and the variety Nancy "partially self-fertile". BRÓZIK—NYÉKI (1975) considered the varieties Szegedi mammut and Ananas to be self-sterile and listed the pollen donors for Szegedi mammut TOMCSÁNYI (1975) and BRÓZIK—SOLTÉSZ (1977) suggested planting pollen donor varieties simultaneously with the varieties Szegedi mammut, Nagykőrösi óriás and Ceglédi óriás in order to avoid problems in the fruit setting. According to NYÚJTÓ (1980) the extent of fertility is satisfactory when every tenth flower sets fruit (10% fruit set).

Self-sterile and "partially self-fertile" varieties give satisfactory yields only when planted in combination with compatible pollen donor varieties which flower simultaneously. In Hungary the flowering time of the apricot varieties currently produced or now being tested were examined in detail by NYÚJTÓ *et al.* (1980a, 1980b), who placed them in three (early, medium and late) flowering categories.

The fertility of free-standing flowers directly determines the volume of yield. BRÓZIK—NYÉKI (1975) and NYÚJTÓ (1978) demonstrated that there is always a greater extent of open pollination than self-pollination. According to BLASSE (1974) and PRISELYUK—ISAKOVA (1977), pollinating self-fertile varieties with the pollen of another variety may also result in an increased rate of fruit set.

Self-sterility may also be traced back to the functional incapability of the sexual organs. One reason for this may be that the female gamete will not accept the sperm of its own variety, while the pollen is viable. This phenomenon was described by SCHULZ (1948) for the apricot



varieties Riland and Perfection, and by BRÓZIK—NYÉKI (1975) for the variety Ananas. The inviability of the pollen may be another cause.

Other, non-genetic characters may also determine self-sterility and "partial self-fertility". EATON—JAMONT (1965), for example, only found gametes capable of functioning in 22% of the flowers. GLUSHKOV—TATAUROVA (1966) and BESPECHALNAYA (1967) found the combined negative effect of high temperatures during dormancy and the ecological conditions of the growing site to be decisive for the extent of fruit set.

MALIGA (1948), NYUJTÓ (1980) and SMYKOV (1978) studied the morphological sterility of the pistil and found that while the pistils of apricot varieties were liable to change in size and morphological characteristics as a function of variety and crop year, 75–100% of them were morphologically fertile in every case.

Under the influence of late frosts disturbances may also occur in the pollen formation, which again has an effect on the extent of fruit set (RYADNOVA 1960).

The investigations on the fruit set of apricot varieties were started in 1954 on the Cegléd and Érd trial grounds of the Research Institute for Fruit Growing and Ornamentals (formerly Horticultural Research Institute), and at Budaörs (Sasad Cooperative Farm), Tiszabura (Lenin Co-operative Farm), Fancsal (Egyetértés Co-operative Farm) and Péntekhely (Balatonboglár State Farm).

Between 1954 and 1978 more than 100 varieties (clones) were examined, of which only those currently produced or recommended for cultivation are discussed in this paper.

The varieties were examined from the following points of view:

Self-fertility — flowers isolated with parchment bags were not pollinated.

Geitonogamy — the isolated flowers were pollinated with pollen collected from flowers of the same variety.

Cross-pollination — flowers isolated and castrated at the white bud stage of flowering were artificially pollinated with pollen another variety.

Pollination with pollen mixture — the isolated and castrated flowers were pollinated with a 1 : 1 mixture of pollen from two different varieties.

Fruit set in free-standing flowers — the amount of fruit developed from unpollinated and artificially pollinated free-standing flowers was examined.

The examinations were carried out according to the method described by MALIGA (1966). The fruit setting percentage was averaged from the total amount of fruit set in 5–35 isolators per combination.

The percentage fruit set was calculated from the amount of fruit developing and ripening from isolated, marked or pollinated flowers (%).

The data were evaluated by the methods of SVÁB (1973).

Fruit set in Hungarian apricot varieties has only been studied by very few authors so far. Of these, MALIGA (1966), BRÓZIK—NYÉKI; (1975) and NYUJTÓ (1978, 1980) made detailed examinations of fruit set after self-pollination, geitonogamy, open and cross pollination.

Owing to the high self-fertility of apricot varieties, the study of this character is not so important as in the case of apple, cherry, sourcherry or almond, for instance, where all or most of the cultivated varieties are self-sterile and must therefore be planted in combination with mutually compatible pollen donor varieties.

According to the results of the investigations, 87.0% (20 varieties) of the major apricot varieties, or of those licensed for propagation, are self-fertile, 8.7% (2 varieties) are "partially self-fertile" and 4.3% (1 variety) is self-sterile (Table 1). No reliable data have so far been obtained on several varieties, e.g. Budapest, Ligeti óriás.

A close examination of self-fertility reveals that the extent of fruit set is greatly influenced by the variety and the crop year (Tables 2–3).

**Table 1**

*Fertility of apricot varieties*  
(1964–1978)

Self-sterile	"Partially self-fertile" Fruit set %	Self-fertile
0.0	0.1–9.9	Above 10
Szegedi mammut	Korai piros (C.242)	Andornaktályai Magyar kajszi
	Korai rózsza (C.508)	Ceglédi bíbor
	Ceglédi óriás	C.265 kajszi
		C.326
		Csongrádi kajszi
		Gönci Magyar kajszi
		Kecskeméti rózsza C.778
		Kécskei rózsza
		Krasnoshchokij Pozdnij
		Magyar kajszi C.235
		C.235/2
		C.256
		C.302
		C.1646
		Pécs 2
	Mandula kajszi C.712	Paksi Magyar kajszi
		Rakovszky kajszi
		Tiszaburai kajszi

**Table 2**

*Self-fertility of apricot varieties*  
(Cegléd, 1978)

Variety	Fruit set, %
Borsi-féle késői rózsza	34.5
Magyar kajszi C.1646	28.5
C.265 kajszi	20.5
Kécskei rózsza	16.7
Gönci Magyar kajszi	11.8
Kecskeméti rózsza C.778	11.1
Mandula kajszi C.712	7.2
Magyar kajszi C.235	5.8
Ceglédi hajnalpír*	5.5
Ceglédi bíbor*	1.9
Nagykőrösi óriás	0.8
Szegedi mammut	0.0
Ceglédi óriás	0.0

\* Spring frost

**Table 3**

*Increase in fruit set in response to artificial self-pollination  
(Budaörs, 1976)*

Variety	Fruit set, %		
	Non-pollinated	Selfed	Surplus fruit set
Ceglédi hajnalpír	6.8	31.2	24.4
Ceglédi óriás	9.6	21.7	12.1
C.326 kajszi	6.8	15.0	8.2
Magyar kajszi C.235	9.3	15.7	6.4
Kecskeméti rózsza C.778	8.9	14.8	5.9

**Table 4**

*Fruit set in isolated non-pollinated, free-standing  
non-pollinated and pollinated flowers  
(Budaörs, 1976)*

Variety	Fruit set, %		
	Isolated non-pollinated	Non-pollinated	Pollinated
Ceglédi óriás	9.6	20.3	17.8
Magyar kajszi C.235	9.3	28.7	18.8
Kecskeméti rózsza C.778	8.9	28.0	25.4
C.326 kajszi	6.8	24.8	22.7
Ceglédi hajnalpír	6.8	4.9	13.2
Average	8.3	21.3	19.6

**Table 5**

*Fruit set in free-standing flowers of apricot varieties  
at various growing sites  
(1978)*

Variety	Growing sites (Fruit set) (%)			Average fruit set, %
	Cegléd	Tiszabura	Péntekhely	
Gönci Magyar kajszi	4.3	24.9	32.3	20.5
Ceglédi bíbor	8.6	16.0	10.9	11.8
Ceglédi óriás	7.4	6.7	15.0	9.7
Nagykőrösi óriás	9.7	8.0	—	8.9



The varieties Ceglédi bíbor and Ceglédi óriás are particularly sensitive to the temperature conditions prevailing at the end of winter and early in spring, since the period of dormancy comes to an end early (NYUJTÓ—BANAI 1975). In response to unfavourable weather conditions the pollen formation may be disturbed, and as a consequence a large proportion of the pollen may become unviable.

There are great differences between the varieties in the extent of fruit set in the same year.

The extent of fruit set resulting from self-pollination may vary from variety to variety. In the present experiments artificial selfing brought about increased fruit setting in each case. Table 3 shows that in 1976 at Budaörs, for example, an 11.4% increase in fruit set was obtained over an average of five varieties.

**Table 6**  
*Fruit set in free-standing flowers in different crop years*  
(Érd)

Variety	Crop year (Fruit set)			Average fruit set, (%)
	1965	1971	1972	
Gönci Magyar kajsz	2	85	59	46.7
Andornaktályai Magyar kajsz	3	90	—	31.0
Nagykőrösi óriás	1	37	11	16.3
Rakovszky kajsz	5	—	52	19.0

**Table 7**  
*Effect of rootstock on fruit set by self- and open pollination*  
*in the variety Magyar kajsz C.235*  
(Cegléd, 1978)

Rootstock	Species	Fruit set, %	
		Self-pollination	Open pollination
C.580	Prunus armeniaca	13	25
Bourdett	Plum	12	17
C.1431	Myrobalan	12	21
C.196	Prunus armeniaca	8	20
C.2546	Prunus armeniaca	8	22
C.303	Prunus armeniaca	7	51
C.174	Myrobalan	6	20
C.2703	Prunus armeniaca	5	13
C.1652	Prunus armeniaca	5	12
C.932	Peach	5	10
C.83	Plum	4	28
C.410	Prunus amigdalo-persica	4	20
Magyar	Prunus armeniaca	4	14
Average		7	21
CV%		43.9	50.0

**Table 8**

*Fruit set after cross-pollination in apricot varieties  
(Cegléd, 1978)*

Pollinated variety	Pollen donors			Average for pollinated variety
	Nagykőrösi óriás	Szegedi mammut	Ceglédi óriás	
Ceglédi bíbor	25.0	15.4	13.3	17.9
Ceglédi óriás	5.2	2.0	2.0*	3.6
Nagykőrösi óriás	0.0*	3.3	3.2	3.3
Szegedi mammut	2.0	0.0*	1.4	1.7
Average of pollen donor variety	10.7	6.9	6.0	6.6

\* Selfing (%)

**Table 9**

*Pollen donors suitable for self-sterile  
and "partially self-fertile" apricot varieties  
(Érd, Cegléd, Budaörs, 1964–1978)*

Pollen donors	Pollinated varieties		
	Szegedi mammut	Nagykőrösi óriás	Korai rózsza (C.508)
Ceglédi bíbor	***	***	**
Ceglédi óriás	**	***	**
Csongrádi kajszai	***	***	—
Gönci Magyar kajszai	***	***	—
Ananász	***	—	—
Ceglédi hajnalpír	—	—	***

\*\*\* Fruit set above 10.0% (good pollen donor)

\*\* Fruit set between 0.1 and 9.9%

— Not examined

The results suggest that the effect on fruit set of artificial self-pollination — carried out by insects and bees under natural conditions — cannot be ignored.

The extent of fruit set in free-standing flowers also proves that open pollination results in a higher rate of fruit set than that observed in the case of isolated, non-pollinated flowers. In the varieties given as examples (Table 4) a 13.1% increase in fruit set was obtained.

Fruit set in free-standing flowers may vary from site to site (Table 5). As seen from the table the varieties give different responses to changes in the ecological conditions (increase or decrease in the rate of fruit set).

The extent of fruit set may be influenced by the crop year (Table 6) and rootstock (Table 7) as well.

Table 10

*Pollination of the apricot variety "Nagykőrösi óriás"  
with a mixture of pollen  
(Cegléd, 1978)*

Pollen donors	Fruit set (%)	Difference (%)				
		1	2	3	4	5
1. Ceglédi bíbor	25.0	—	8.1	15.3	19.8	24.2
2. Ceglédi bíbor + Ceglédi óriás	16.9	—	—	7.2	11.7	16.1
3. After pollination	9.7	—	—	—	4.5	8.9
4. Ceglédi óriás	5.2			—	—	4.4
5. Selfing	0.8					—

The effect of cross-pollination was studied on the self-sterile variety Szegedi mammut and on "partially self-sterile" varieties. The results show that the apricot varieties Ceglédi óriás, Nagykőrösi óriás és Szegedi mammut are not suitable for use as mutual pollen donors, due probably to the low viability of the pollen (Table 8). An increased rate of fruit set was observed in each case when using the pollen of the variety Ceglédi bíbor.

On the basis of the examinations performed so far, Table 9 lists the suitable pollen donors for self-sterile and "partially self-fertile" apricot varieties. The table reveals that the varieties Ceglédi bíbor and Ceglédi óriás produced an adequate rate of fruit set in certain crop years, which supports the results of investigations made by NYÚJTÓ—BANAI (1975).

The effect of a pollen mixture on fruit set is shown in Table 10. In the variety Nagykőrösi óriás the extent of fruit set obtained with a pollen mixture was lower than that obtained with the pollen of the variety Ceglédi bíbor.

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### References

- BESPECHALNAYA, V. V. (1967): Osobennosti cvetenija i zavjazüvanija plodov abrikosa v uslovijah Moldavii. Sadov. Vinogr. Vinod., **22**, 14–17.
- BLASSE, W. (1974): Scherung der Bestäubung industriemässigen obstanlagen. Gartenbau-wiss., **21**, 4, 113–115.
- BRÓZIK, S.—NYÉKI, J. (1975): A kajszai termékenyülési viszonyai. [In: S. Brózik, J. Nyéki (szerk.): Gyümölcstermő növények termékenyülése.] [Fruit setting in apricot. In: S. Brózik, J. Nyéki (eds): Fruit set in fruit-bearing plants.] Mezőgazdasági Kiadó, Budapest. 173–176.
- BRÓZIK, S.—SOLTÉSZ, M. (1977): Kajszai. [In: F. Gyuró (szerk.): Gyümölcsfajták társítása.] [Apricot. In: F. Gyuró (ed.): Association of fruit varieties.] Mezőgazdasági Kiadó, Budapest. Ma újdonság — holnap gyakorlat 58.
- EATON, G. W.—JAMONT, A. M. (1965): Embryo sac development in the apricot, *Prunus armeniaca* L. Constant. Amer. Soc. Hort. Sci., **86**, 95–101.



- GLUSHKOV, A. I.—TATAUROVA, A. S. (1966): Nekatoruje osobennosti biologii cvetenija sortov abrikosa v uslovijah Srednej Azii. Sborn. Trud. Asp. Moldav. Nauchn. Szotr. 7, 395–402.
- HORN, J. (1939): Kajszi-, cseresznye-, meggytermesztés (The production of apricots, cherries and sour cherries). Növényvédelem és Kertészet Kiadása, Budapest.
- MALIGA, P. (1948): Adatok a kajszi- és kajszi-alkati meddőségéhez (Morphological sterility of apricot trees). Agrártud. Egyetem Kert- és Szőlőgazd. Tud. Kar Közlem., 12, 1–7.
- MALIGA, P. (1966): Kajszi-alkati termésnyúlási viszonyai (Fruit setting in apricot varieties). Szőlő- és Gyümölcsterm., 1, 87–97.
- NYÚJTÓ, F. (1978): A kajszi-alkati ültetvényekben alkalmazható fajták ismertetése. (In: A csonthéjas gyümölcsűek fajtái, termesztéstechnológiája és a betakarítás lehetőségei. Újabb kutatási eredmények a gyümölcstermesztésben.) [Varieties suitable for use in apricot orchards. (In: Varieties and production technologies of stone fruits, and possibilities of harvesting. Recent research results in fruit growing.)] Gyümölcs- és Dísznövénytermesztési Kut. Int. Kiadványa, Budapest, 23–31.
- NYÚJTÓ, F. (1980): Kajszi-alkati. (In: J. Nyéki (szerk.): Gyümölcsfajták virágzásbiológiája és termésnyúlása.) [Apricot. In: J. Nyéki (ed.): Flowering biology and fruit set in fruit varieties.] Mezőgazdasági Kiadó, Budapest, 248–262.
- NYÚJTÓ, F.—BANAI, B. (1975): Előzetes közlemény a kajszi-alkati fajták téli morfológiájának vizsgálatáról (Preliminary report of studies on the winter morphology of fruit buds in apricot varieties). Gyümölcsterm., 2, 15–21.
- NYÚJTÓ, F.—BRÓZIK, S.—NYÉKI, J.—BRÓZIK, S. JR. (1980a): Kajszi-alkati fajták társítása (Association of apricot varieties). Kertgazdaság, 12/3, 23–32.
- NYÚJTÓ, F.—BRÓZIK, S.—NYÉKI, J.—BRÓZIK, S. JR. (1980b): Kajszi-alkati fajták virágzása (Flowering of apricot trees). Acta Agron. Hung., (in press).
- PRISELNYUK, G. T.—ISAKOVA, M. D. (1977): Stepen' samopodosti i podbor opülitelej dlja abrikosa. Sadov. Vignor. Vinod. Mold., 12, 18–19.
- RYADNOVA, I. M. (1960): Vlijanie temperaturnyh uslovij na razvite cvetocnyh pocshok i plodov. Fiziol. Rast., 7, 92–94.
- SCHULTZ, J. H. (1948): Self-incompatibility in apricots. Proc. Amer. Soc. Hort. Sci., 51, 171–174.
- SMYKOV, V. K. (1978): Biologija jabloni i abrikosa i principu formirovanija promüshlennih sortimentov. Stiinca, Kisinev.
- SVÁB, J. (1973): Biometriaei módszerek a kutatásban (Biometric methods in research). Mezőgazdasági Kiadó, Budapest.
- TOMCSÁNYI, P. (ed.) (1975): Információk a gyümölcsfajtákról. Ma újdonság — holnap gyakorlat (Information on fruit varieties. Today a novelty — tomorrow common practice). Mezőgazdasági Kiadó, Budapest.

## EFFECT OF MALUS POLLINATORS ON THE QUALITY OF APPLE

The possibilities for using *Malus* species and types (wild and ornamental apples) as pollinators have been studied in Hungary since 1977 (NYÉKI *et al.* 1981). The introduction of the "Malus pollination system" enables the establishment of single-variety apple orchards even in the case of self-sterile varieties (GYURÓ *et al.* 1980). Besides the characteristics of flowering phenology, fruit setting biology and growth, the quality of the fruit in the pollinated varieties must also be taken into consideration when choosing the right species or types of *Malus* pollinators (SOLTÉSZ *et al.* 1979). It is highly important to ensure that the *Malus* species and types used as pollinators do not cause unfavourable metaxenic changes in the (♀) fruits of commodity varieties.

Using *Malus baccata* as pollinator, NEBEL—KERTÉSZ (1934) obtained a higher sugar content and darker juice colour, while the skin colour and fruit shape did not change. According to observations by English authors (ANONYMOUS 1972) certain *Malus* species may cause increased suberization. WILLIAMS—CHURCH (1974) found that *Malus* pollinators did not reduce the size of the fruit. JONKERS *et al.* (1978) reported on positive changes over an average of three years.

The *Malus* species and types used as pollinators in the present experiment were obtained from the Botanical Garden of the University of Horticulture (Soroksár). The types, marked with the SBK code numbers of the Botanical Garden, were chosen from the species *Malus silvestris*, *spectabilis*, *floribunda*, *baccata*, *pumila*, *dasyphylla* and *halliana*. The pollen gathered was applied to the flowers of commodity varieties (Jonathan, Starking, Golden Delicious and Staymared) at the Helvécia Station of the National Institute for Agricultural Variety Trials. In each combination 150–200 flowers were pollinated. For the sake of comparison the test varieties were pollinated with each other's pollen, and fruits obtained from open pollination were also examined.

The fruit size was measured every year from 1977 to 1980. A slide-gauge was used to measure the largest diameter and length of the fruit to a precision of one millimetre. The "shape index" of the fruit was calculated from the ratio of length to diameter.

In 1980 an informatory examination of the redness of the peel and the physical and internal quality characteristics of the fruits was carried out. The fruits of varieties with red peel were divided into four groups according to the degree of redness on the fruit surface. The four groups were: 0–25; 25.1–50; 50.1–75 and 75.1–100 per cent red surface.

In the course of laboratory examinations the basic colour was determined with the aid of a colour scale, the firmness of the flesh by means of a hand penetrometer, the state of starch decomposition on the basis of discoloration after dipping in potassium iodide solution; the soluble dry matter content was established with a hand refractometer, the sugar content by the Luff—Schoorl method, while the acid content was measured by titration with 0.1 N NaOH. The pomona value (total internal quality value) was calculated by totalling the tenfold values of sugar (g/l) and acid (g/l) content.

Table 1 shows the effect of *Malus* pollinators on the diameter of the fruit, the most important of all the data from a commercial point of view. The effect of the type of pollinator changed with the variety, and in certain combinations even with the year. The standard deviation was in most cases 0.2–0.4. It was only in certain years that the Jonathan fruits (♀) showed a reduction in size under the influence of certain pollinators; in other years this phenomenon was not observed. The pollination of Starking (♀) resulted in ripe fruits in less combinations, because in this variety many fruit primordia were shed in response to low temperatures after fruit setting. It was also in this variety that changes in size occurred most frequently; the fruits were usually smaller than in the control. The Golden Delicious fruits (♀) were generally more uniform in size. A significant reduction in size was obtained on two occasions with the types SBK-333 and 1003 (♂).

Owing to opposing changes in the length and diameter the "shape index" (length/diameter) of the fruit was sometimes modified. In comparison with fruits obtained by open pollination the *Malus* types SBK-9 and 10 decreased the shape indexes of Jonathan, Golden Delicious and Starking fruits every year. The latter two varieties generally responded with a reduced "shape index" to other pollinators too.

Certain *Malus* types also changed the colouration of the fruit (Table 2). In the case of Jonathan (♀) the type SBK-8 brought about a far better fruit colouration compared to those obtained by open pollination; not only the extent but also the intensity of the red colour increased. In the variety Staymared (♀) a fruit colour better than in the control was attained with two pollinator types, SBK-8 and 9, while fruits originating from the pollinator SBK-7 showed very poor colouration. In Starking combinations no significant modification was observed. In the variety Golden Delicious 10% of fruits reaching the state of ripeness displayed a characteristic "metaxenic redness" in response to pollination by SBK-1 and 10.

The results of laboratory analyses are presented in Tables 3, 4 and 5. The flesh firmness, an important factor in transportability, increased under the influence of *Malus* pollinators. The basic colour and the analytical results for starch decomposition are not indi-



Table 1

*Effect of Malus species (types) as pollinators on the diameter of Jonathan, Starking, Golden Delicious and Staymared fruits (mm) (Hélvécia)*

Code numbers of Malus types (♂)	Jonathan (♀)				Starking (♀)				Golden Delicious (♀)				Stay- mared (♀)
	1977	1978	1979	1980	1977	1978	1979	1980	1977	1978	1979	1980	1980
SBK-1	56	57		67	64	54			56	57	57	64	55
SBK-3	62	59			53				65	62	61		
SBK-5	52	57			45				65	56	60		
SBK-6	51	55	73		59		66		65	59	63		
SBK-7	53	69		66	52						64	59	58
SBK-8		70		67				61		60		68	55
SBK-9	58	61	73	72	51	53				63	54	63	55
SBK-10	63	57		67	59	50			58	60	67	64	59
SBK-13				71								60	
SBK-15				73								64	57
SBK-277	53		70		63				59		59		
SBK-333	51		68		65				54		54		
SBK-336	60	58	70		54	51			62	57	63		
SBK-726	53		69		65				64				
SBK-730	51		73		57				59		59		
SBK-1003	64	66	72		58				58	55			
SBK-1014	60	65	68		61		79		59	61	58		
Jonathan					52	62	66	70	65	62	66		
Starking	68	63	67						68	61	54		
Golden Delicious	61	60	67		56	63	70						
Open pollination	60	62	69	70	63	63	72	71	62	57	59	68	59

Table 2

*Effect of Malus species (types) on the extent of colouring of the fruit surface (Hélvécia, 1980)*

Pollinator Malus type (♂)	Jonathan (♀)				Staymared (♀)			
	0-25	25.1-50	50.1-75	75.1-100	0-25	25.1-50	50.1-75	75.1-100
	per cent of fruit surface covered by red colour							
SBK-1	66	27	7	0	47	29	24	0
SBK-7	43	43	0	14	100	0	0	0
SBK-8	0	12	38	50	0	17	83	0
SBK-9	69	10	17	4	38	12	25	25
SBK-10	45	28	24	3	17	50	33	0
SBK-13	61	33	6	0				
SBK-15	42	33	17	8	0	75	25	0
Open pollination	38	45	12	5	14	47	39	0



Table 3

*Effect of Malus species (types) as pollinators on the internal quality of Jonathan apples (♀) as determined by laboratory analysis (15 September 1980)*

Type of pollinator (♂)	Basic colour (scale)	Firmness of flesh, kg/cm <sup>2</sup>	Starch (scale)	Refrac-tion, %	Sugar content, %			Acid content (malic acid%)	Sugar to acid ratio	Pomona value
					total	reduc-ing	saccha-rose			
SBK-1	5.9	8.26	2.2	13.6	12.26	8.80	3.48	1.105	11.1	233.1
SBK-7	5.9	8.33	1.9	14.8	13.26	9.43	3.83	1.260	10.5	258.6
SBK-8	6.0	8.60	1.9	15.2	13.36	9.84	3.52	1.195	11.1	253.1
SBK-9	5.8	7.76	2.3	13.2	11.96	8.02	3.94	1.285	9.3	248.1
SBK-10	6.3	8.24	2.2	13.2	12.06	8.60	3.46	1.215	9.9	242.1
SBK-13	5.9	8.09	1.8	14.0	12.66	8.75	3.91	1.155	11.0	242.1
SBK-15	6.0	8.20	1.8	13.8	12.46	8.60	3.86	1.200	10.4	244.6
Open pollination	5.9	7.98	2.3	14.4	12.86	8.96	3.90	1.135	11.3	255.0

Table 4

*Effect of Malus species (types) as pollinators on the internal quality of Staymared apples (♀) as determined by laboratory analysis (30 September 1980)*

Type of pollinator (♂)	Basic colour (scale)	Firmness of flesh, kg/cm <sup>2</sup>	Starch (scale)	Refrac-tion, %	Sugar content, %			Acid content (malic acid%)	pH	Sugar to acid ratio	Pomona value
					total	reduc-ing	saccha-rose				
SBK-1	4.9	10.42	1.6	13.0	11.87	8.70	3.17	0.760	3.25	15.6	194.7
SBK-7	5.5	9.92	1.5	12.0	10.12	7.57	2.55	0.827	3.18	12.2	183.9
SBK-8	4.7	10.57	1.7	13.0	11.67	8.76	2.91	0.824	3.20	14.16	199.1
SBK-9	4.7	10.79	2.1	13.4	11.56	10.47	1.09	0.744	3.40	15.62	190.0
SBK-10	5.6	19.95	1.5	13.6	13.50	8.07	5.43	0.797	3.04	16.9	214.7
SBK-15	4.0	10.58	1.7	13.8	12.26	8.96	3.30	0.864	3.27	14.3	209.0
Open pol-lination	5.0	9.71	1.5	13.2	12.36	8.49	3.87	0.797	3.20	15.5	203.3

cative of any difference in ripeness, i.e. the increased flesh firmness was independent of the state of ripening.

Other metaxenic changes were also observed. In 1980 10–20% of the Jonathan fruits obtained by pollination with SBK-1, 9 and 15 had open calyces, and all Jonathan fruits originating from the pollinator SBK-10 had open calyces. In the same variety types SBK-1 and 9 caused a shortening of the stem and a thickening of the stem end in 10–15% of the fruit. In Golden Delicious fruits (♀) an 8–18% occurrence of very long (4 cm or longer) stems was found as a consequence of pollination with SBK-1, 7, 10 and 15. In the case of Staymared (♀) the pollinators SBK-1 and 9 brought about similar changes 17–37% of cases.

The results show that under continental climatic conditions similar to those in Hungary the occurrence of metaxenia must be reckoned with, and this fact must be taken into

Table 5

*Effect of Malus species (types) as pollinators on the internal quality of Golden Delicious apples (♀) as determined by laboratory analysis (30 September 1980)*

Type of pollinator (♂)	Basic colour (scale)	Firmness of flesh, kg/cm <sup>2</sup>	Starch (scale)	Refracti-on, %	Sugar content, %			Acid content (malic acid%)	pH	Sugar to acid ratio	Pomona value
					total	reduc-ing	saccha-rose				
SBK-1	4.9	9.49	3.0	14.4	13.92	9.03	4.89	0.717	3.28	19.3	210.9
SBK-7	4.4	9.35	4.0	14.6	13.63	9.79	3.84	0.650	3.48	21.0	201.3
SBK-8	5.0	8.89	3.7	15.2	14.30	10.52	3.87	0.640	3.30	22.5	207.0
SBK-9	4.5	9.55	3.2	14.7	13.59	10.10	3.49	0.778	3.28	17.4	213.7
SBK-10	5.0	9.43	2.5	14.0	14.18	9.55	4.63	0.766	3.33	18.9	218.4
SBK-13	5.0	9.86	3.6	15.3	14.39	10.00	4.39	0.611	3.27	23.6	205.0
SBK-15	4.9	9.04	3.2	15.0	14.39	10.31	4.08	0.737	3.30	19.4	217.3
Open pollina-tion	4.5	8.28	3.4	14.4	13.89	9.03	4.86	0.777	3.28	17.8	216.6

consideration when deciding on the applicability of *Malus* species and types as pollinators. In the types giving the best results from the points of view of flowering and fruit-setting biology, observations on metaxenia will be made on a larger number of fruits in the following years.

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### References

- ANONYMOUS (1972): Pollination of fruits. Report 1971. Long Ashton Research Station, Arrowsmith Ltd., Bristol. 24-28.
- GYURÓ, F.—TERPÓ, A.—NYÉKI, J.—SOLTÉSZ, M.—TÓTH, M. (1980): Fajtatizsza gyümölcs-ültetvények létesítése, mint a műszaki fejlesztés alapja (Establishment of single-variety orchards, as a basis for technical development). Lippay János Tudományos Ülésszak, Kertészeti Egyetem, Budapest (in press).
- JONKERS, H.—BORSBOOM, O.—HOLDER, V. (1978): Fécondation du pommier par des pommiers ornementaux. Groupe de travail "Cultures horticoles". Ecole supérieure agronomique. Wageningen. Le Fruit Belge. Bulletin trimestriel, 46/331, 67-75.
- NEBEL, F.—KERTÉSZ, F. (1934): Metaxenia and xenia in apples. Gartenbau, 9, 45-64.
- NYÉKI, J.—SOLTÉSZ, M.—TÓTH, M.—GYURÓ, F.—TERPÓ, A. (1981): Fajtatizsza almaültetvények megporzási lehetőségei *Malus* fajokkal (Possibilities of using *Malus* species as pollinators in single-variety apple orchards). Acta Agron. Hung., (in press).
- SOLTÉSZ, M.—GYURÓ, F.—TERPÓ, A.—NYÉKI, J.—TÓTH, M. (1979): Üzemi almafajták és *Malus* fajok társításának lehetőségei (Possibilities of combining commercial apple varieties with *Malus* species). Kertgazdaság, 11/5, 1-16.
- WILLIAMS, R. R.—CHURCH, R. (1974): Can *Malus* pollinators do this to your apples? Grower, 81/16, 770, 772.



## EFFECT OF INBREEDING ON SOME MAJOR CHARACTERISTICS OF HEMP

Inbreeding in hemp is a question which has been studied very little so far. The reason for this is that dioecy makes self-pollination impossible, while sister line crossing in spatial isolation takes a long time and meets with technical difficulties. In the case of monoecious hems capable of self-pollination, on the other hand, the effect of inbreeding has not been studied at all, partly because, owing to its large size, the plant is difficult to isolate artificially, and does not readily tolerate artificial isolators. Individual flowering under conditions of spatial isolation is therefore the only solution, which in itself restricts the number of self-pollinated plants.

Since, however, heterosis breeding has become general practice for cross-pollinated plants, the production of inbred hybrids is desirable in the case of hemp, too. Thus, besides continuing the work of FLEISCHMANN (1934, 1941) and BÓCSA (1958) on dioecious hemp, inbreeding on monoecious hemp has been started from scratch.

The first literary data on inbreeding in hemp were published by FRUWIRTH (1922), who did not describe the method applied, but noted that inbreeding had no effect whatsoever on the length of the stem or on the morphological characters. In his opinion many  $S_2$ - $S_3$  generations were superior to the initial material as regards technical and absolute stem length. BÓCSA (1958) found in his experiments that the stem length of inbred "F" hemp was 68% of that of the initial material. In the inbred lines produced by STEPANOV (1975) the proportion of fertile pollen substantially decreased. In the course of his investigations he found the proportion of fertile pollen in monoecious hemp (variety: Juszo-1) to be 100% compared to an average of 4.1% in  $S_1$ , 3.1% in  $S_2$ , 1.5% in  $S_3$  and 1.5% in  $S_4$ . Similarly low values were obtained with dioecious hems. The latter values are so conspicuously low that they can safely be regarded as those pertaining to male sterile forms. Fertile pollen proportions as low as these are not mentioned in the literature as occurring in other species. According to BÓCSA *et al.* (1974) the proportion of fertile pollen in lucerne decreased from  $S_0$  to  $S_4$  by a mere 18.3%. FLEISCHMANN (1934) reported a 50% reduction in seed production during some 15 years of inbreeding.

According to the investigations of FLEISCHMANN (1941) into the production potential of a hemp line inbred for 15 years the stem yield showed a sharp reduction compared with  $S_0$  in response to inbreeding over a long period of time.

On studying the stem production in various monoecious lines segregated from a dioecious inbred hemp, BÓCSA (1958) found that the depression was also manifested in a great decrease in stem yield. It is characteristic of the extent of depression that the stem yield of the most productive inbred line included in the experiment was 35% lower and that of the poorest line 47% lower than that of the non-inbred dioecious control Kompolti.

The material used for the production of dioecious inbred hems was a plant stock called Fleischmann Inbred (hereinafter: F. Inb.) which had been used for loose inbreeding between 1919 and 1951. This means that on a spatially isolated area the seed of a single mother plant selected from the same population was sown every year. In 1951 this material was divided into strains by Bócsa, who maintained 18 strains by spatial isolation, but only a single male plant per strain was allowed to flower, and the seed of a single female hemp was sown in each place. Thus, up till now sister line breeding has been taking place. Inbreeding, in the strictest sense of the word, only really began in 1952.

Inbred material from Tiborszállás (hereinafter called T. Inb.) was transferred to Kompolt from the Cereal Research Institute, Szeged, in 1972. Its origin can be traced back to 1965 or 1968, like that of the Szeged inbred material (S. Inb.) which was also taken to Kompolt in 1972 when hemp breeding was discontinued at Szeged. An essential feature of the method of inbreeding is that the male partner marked out for pollination and the female partner to be propagated are phenotypically the most developed plants. The inbred strains were used to set up a densely-sown fibre experiment and a wide-spaced seed hemp experiment to determine their stem and seed production potentials.

The monoecious inbred strains were produced from Fibrimon 21-63, a French monoecious variety maintained at Kompolt. From this variety 6 mother plants possessing good phenotypic characteristics were chosen in 1977. In order to produce monoecious inbred strains the winter period also had to be utilized, with intensive photoperiodic treatments in the greenhouse, so that by mid-April at the latest mature seed was obtained for sowing outdoors and producing the next inbred generation. Monoecious hems were sown isolated in maize stands. Part of the self-pollinated seed of the mother plant was sown the following year at wide spacing, while the rest was further propagated as a sib, since seed produced by a single mother plant would not have been sufficient for a comparative experiment of fibre hemp. (Some seed was, of course, retained to raise a plant for further self-pollination.) For technical



reasons only the  $S_2$  generation could be used in fibre hemp trials, while at a wide spacing the  $S_1$ ,  $S_2$  and  $S_3$  generations were used, although  $S_2$ , for example, did not have quite the same genetic value, because, for technical reasons, a seed mixture of sister lines from the same generation, or sib seed derived from intercrossing had to be used in the fibre hemp experiment. Thus, there was a considerable difference in the degree of inbreeding between the original mother plant and the sibs from the same generation.

In the course of growth and development, physiological and agronomical characters were examined in both seed and fibre hemp experiments. In each plot 10–20 plants were cut and dried, after which the height, thickness and weight were recorded.

The fertility of the pollen was also studied. The pollen quality of inbred hemp was characterized by the size and proportion of fertile and sterile pollen. The examinations were performed with an NFPK microscope at  $400\times$ . The flowers removed were stored at room temperature until use. The pollen was stained using the method of ALEXANDER (1969). The ratio of fertile to sterile pollen was determined in 30 fields of sight per variety.

In order to study the effect of inbreeding on agronomical characters a comparative trial was laid out to determine the stem production potential. In 1978–1980 monoecious and dioecious inbred generations were sown together with the initial varieties in comparative fibre and seed production trials in a random block design with 4–5 replications. The size of plot was 7.7–8 m<sup>2</sup> for fibre hemp and 10–40 m<sup>2</sup> for seed hemp. For fibre hemp the germ num-

**Table 1**  
*Effect of inbreeding on the major characters of hemp stems*

Variety	Height		Thickness		Weight	
	cm	%	cm	%	g	%
densely sown						
Kompolti	235.7	100.0	8.5	100.0	17.3	100.00
S. Inb.	218.9	92.87	7.7	90.59	11.9	65.79
T. Inb.	204.1	86.59	6.9	81.18	9.6	55.49
F. Inb.	174.0	74.82	6.0	70.59	7.3	42.20
SD5%	8.1	—	0.6	—	1.3	
wide-spaced						
Kompolti	391.2	100.00	28.6	100.00	391.5	100.00
S. Inb.	347.7	88.88	27.0	94.41	331.2	84.60
T. Inb.	323.5	82.67	24.9	87.06	297.3	75.94
F. Inb.	293.2	74.93	24.1	84.27	208.9	53.36
SD5%	16.5	—	1.5	—	36.5	
wide-spaced						
Fibr. $S_0$	288.44	100.00	24.04	100.00	296.60	100.00
Fibr. $S_1$	248.44	86.13	21.20	88.19	245.46	82.76
Fibr. $S_2$	211.36	73.28	17.82	74.13	206.64	69.67
Fibr. $S_3$	190.66	66.10	15.04	62.56	183.00	61.70
SD5%	10.59	—	1.87	—	6.98	—

ber was 4.5 million/ha. Harvesting took place at the stage of technical maturity, which means 50% flowering of male hems in dioecious varieties and 10% flowering of male flowers in monoecious varieties. In the fibre hemp trials, after determining the dry stem yield, samples of 6–10 kg per plot and per variety were taken for the analysis of components; after retting, and mechanically extracting the fibre the fibre yield was determined.

The seed production trials were sown every year with a row space of 70 cm and a plant distance of 60 cm. Besides the yield the major yield components were also measured.

The stem, fibre and seed yields and the yield components of the seed were evaluated by variance analysis. Finally, the coefficients of inbreeding were calculated using the formula  $F = 1 - \left(1 - \frac{1}{2N}\right)^n$  based on a population size of  $N_e = 2$  for dioecious and  $N_e = 1$  for monoecious hems (Malécot 1948 in Sváb 1971).

#### 1. Effect of inbreeding on the major characters of the hemp stem

In the course of growth and development in the wide spaced F. Inb. strain the height, thickness and weight of the stem reached only 74.93, 84.27 and 53.36% respectively of the corresponding parameters in the non-inbred initial material. In a dense stand the stem measurements of the inbred dioecious hemp were reduced at a faster rate than at a wide spacing (Table 1). The stem measurements decreased to a greater extent in inbred male hems than in inbred female hems (Table 2).

**Table 2**  
*Effect of inbreeding on the stem yield of hemp when sown densely and wide spaced*

Variety and strain	Fibre hemp		Seed hemp	
	kg/plot	%	kg/plot	%
1978 Male plants				
Kompolti	9.04	100.00	14.4	100.00
S. Inb.	7.73	85.51	11.68	82.60
T. Inb.	7.22	79.83	9.27	65.57
F. Inb.	6.30	69.66	6.96	49.22
SD5%	1.34	15.09	2.09	11.98
1979 Female plants				
Kompolti	10.78	100.00	28.71	100.00
S. Inb.	9.77	90.63	23.43	91.13
T. Inb.	8.31	77.09	21.80	83.23
F. Inb.	6.71	62.21	14.42	56.08
SD5%	0.78	8.18	1.91	6.25
1980 Monoecious				
Kompolti	9.44	100.00	—	—
Fibr. S <sub>0</sub>	8.53	100.00	8.12	100.00
Fibr. S <sub>1</sub>	—	—	6.47	79.69
Fibr. S <sub>2</sub>	6.86	72.66	5.08	62.56
Fibr. S <sub>3</sub>	—	—	3.47	42.74
SD5%	1.09	14.53	0.55	5.91

In self-pollinated monoecious strains the average height of  $S_3$  plants was 66.1%, the stem thickness 62.56% and the stem weight 61.7% compared to the corresponding values in the original variety. With respect to the characters examined, the order was: S. Inb. > T. Inb. > F. Inb. for dioecious hemp and  $S_1 > S_2 > S_3$  for monoecious hemp. Inbreeding may cause changes in the number and length of the internodes. In the dioecious and monoecious inbred stands in general the internodes in the vegetative part of the plant increased in number but decreased in length.

## 2. Effect of inbreeding on hemp pollen

As a response to inbreeding the ratio of fertile to sterile pollen changed. In the dioecious inbred strains S., T. and F. the proportion of fertile pollen is some 6, 10 and 12% lower, respectively, than in the non-inbred Kompolti (Table 3). In the monoecious inbred strains the proportion of fertile pollen decreased by 11.44% from  $S_0$  to  $S_3$  (Table 3). In fact, it took three inbred generations for monoecious hemp to fall to the pollen sterility level of dioecious inbred varieties in the  $S_{27}$  (F. Inb.) or  $S_{15-13}$  (S. Inb. and T. Inb.). The relatively small difference in pollen sterility between  $S_{13-15}$  and  $S_{27}$  seems to prove that reduction below the 85% level of pollen fertility is a very slow process, and that this value is practically the minimum.

Inbreeding results in changes not only in the ratio of fertile to sterile pollen but in the dimensions of the pollen too. In the dioecious F. Inb. the length and breadth of fertile pollen were 81.63 and 82.55% of the respective measurements for fertile pollen in non-inbred hems (Table 3).

**Table 3**  
*Effect of inbreeding on the stem yield  
of hemp when sown densely and wide spaced  
and on the dimensions of the pollen*

Variety and strain	Pollen, %		Length of pollen		Breadth of pollen	
	fertile	sterile	$\mu$	%	$\mu$	%
Kompolti	95.43 $\pm$ 3.28	4.57 $\pm$ 3.30	28.60 $\pm$ 0.53	100.00	27.47 $\pm$ 0.47	100.00
S. Inb.	87.88 $\pm$ 6.67	12.12 $\pm$ 6.67	24.21 $\pm$ 0.69	84.65	23.58 $\pm$ 0.71	85.84
T. Inb.	85.71 $\pm$ 3.37	14.29 $\pm$ 3.77	24.01 $\pm$ 0.65	83.95	22.67 $\pm$ 0.65	82.53
F. Inb.	81.61 $\pm$ 6.89	18.39 $\pm$ 2.25	23.61 $\pm$ 0.41	82.55	22.42 $\pm$ 0.45	81.63
Fibr. $S_0$	95.62 $\pm$ 1.40	4.38 $\pm$ 1.40	27.89 $\pm$ 0.49	100.00	26.80 $\pm$ 0.43	100.00
Fibr. $S_1$	89.70 $\pm$ 3.26	10.30 $\pm$ 3.21	24.18 $\pm$ 0.55	86.70	23.41 $\pm$ 0.57	87.35
Fibr. $S_2$	85.92 $\pm$ 1.47	14.08 $\pm$ 1.47	23.60 $\pm$ 0.43	84.62	22.55 $\pm$ 0.57	84.14
Fibr. $S_3$	83.99 $\pm$ 1.88	16.01 $\pm$ 2.49	22.53 $\pm$ 0.51	80.78	21.63 $\pm$ 0.53	80.71

After three years of self-pollination the length and breadth of fertile pollen in monoecious  $S_3$  plants were reduced by 19.22 and 19.29%, respectively. With respect to the inbreeding minima of pollen measurements the same is true as in the case of fertility. The order of varieties and generations is exactly the same as for the stem properties.

## 3. Effect of inbreeding on the agronomical characters of hemp

(a) *Stem yield.* In inbred strains sown densely the proportion of inferior plants (not higher than 50% of the average technical stem length of the stand) considerably increased. The trend was as follows:

Kompolti	100.0
Fibrimon	140.6
S. Inb.	162.9
T. Inb.	175.9
F. Inb.	218.7

As seen above, in the material inbred for the longest period the proportion of inferior plants in the stand increased twofold suggesting a sharp decline in vigour. The order is the same again.



As regards the stem yield, both the male and female plants of dioecious hems show a considerable depression. F. Inb. had reached the inbreeding minimum, because in comparison to Bócsa's earlier experiments (unpublished) its stem yield showed no further decrease. The order is again Kompolti > S. Inb. > T. Inb. The male plants showed a greater depression than the female plants, which demonstrates the well-known fact that male hems are less robust and vigorous in every respect than female hems. It is worth noting that in monoecious hemp a 28% depression had occurred by the  $S_2$ . On the other hand, in the case of monoecious hems the great difference in stem yield between fibre and seed hems, and the extremely low value in  $S_3$  can be attributed to the fact that the degree of inbreeding is not uniform. As mentioned above, seed hemp is always identical with the original self-pollinated mother plant, while fibre hemp is its sibbed progeny. The inbreeding coefficient of the latter is considerably lower.

(b) *Fibre content.* In dioecious hems particularly, inbreeding caused a considerable reduction in fibre content, although it is very difficult to judge the original fibre content of the initial material, as it is certain that there were very great differences between the varieties in this respect. For example, S. Inb. originated from the variety Szegedi 9, which had a high fibre content, and this made its effect felt even after 13 inbred generations. Also, the dif-

**Table 4**  
*Effect of inbreeding on fibre content in dioecious and monoecious hems*

Variety and strain	Fibre content		
	wide-spaced		densely sown, %
	male, %	female, %	
1978			
Kompolti	28.4	32.1	35.8
S. Inb.	—	—	24.7
T. Inb.	—	—	19.8
F. Inb.	11.8	11.3	13.0
1979			
Kompolti	29.0	33.3	33.9
S. Inb.	13.6	16.1	19.5
T. Inb.	12.4	14.0	16.2
F. Inb.	11.3	12.9	13.0
1980			
Fibr. S <sub>0</sub>	—	—	31.1
Fibr. S <sub>2</sub> -83	—	—	28.6
Fibr. S <sub>2</sub> -85	—	—	27.1
Fibr. S <sub>2</sub> -87	—	—	29.0
Fibr. S <sub>2</sub> -88	—	—	29.6
Fibr. S <sub>0</sub>	23.8		
Fibr. S <sub>1</sub>	22.1		
Fibr. S <sub>2</sub>	21.3		
Fibr. S <sub>3</sub>	20.2		

ference in fibre content between densely-sown and wide-spaced stands, as well as between male and female plants, is similarly maintained. It is worth mentioning that the fibre content decreased to an even greater extent than the stem yield in response to inbreeding; in F. Inb. it was only 38–45% of the standard value for Kompolti. In this case selection complicates the situation, since the variety Kompolti has been under intensive selection for fibre content for nearly 30 years. It is therefore difficult to compare the inbred and selected (control) varieties from this point of view.

In monoecious hems the fibre content becomes substantially lower even after two generations, as seen in the wide-spaced hemp (true  $S_2$  generation) (Table 4). The sib- $S_2$  generations, on the other hand, showed no particular depression, quite understandably considering their low inbreeding coefficients. The effect of inbreeding on the fibre content is best seen in the wide-spaced monoecious strains where the results are not distorted by selection for fibre content. The effect of selection for fibre content during 3 generations is negligible.

(c) *Seed production.* Seed yield and its major components were examined in each plot. It is well known that the number of seeds per plant and the thousand-seed-weight jointly

**Table 5**  
*Effect of inbreeding on seed yield and its components*

Variety and strain	Seed yield		Number of seed		Thousand-seed-weight	
	kg/plot	%	n/plot	%	g	%
Kompolti	3.78	100.0	3271	100.0	19.1	100.0
S. Inb.	3.38	89.4	3189	94.6	19.0	99.3
T. Inb.	2.90	76.7	2946	87.4	18.4	96.6
F. Inb.	2.51	66.4	2900	86.0	16.0	83.7
SD5%	0.34	9.71	171			
Fibr. $S_0$	2.08	100.0	6645	100.0	15.2	100.0
Fibr. $S_1$	1.27	61.0	4485	67.5	14.3	93.9
Fibr. $S_2$	0.94	45.2	3553	53.5	13.3	87.5
Fibr. $S_3$	0.81	38.9	3322	50.0	12.7	83.2
SD5%	0.13	6.3	767			

determine the seed yield. The values of these two yield components combine to form the seed yield. It can be established beyond any doubt that of all the factors examined seed production showed the greatest decrease in response to inbreeding. It is extremely interesting that in spite of the long period of inbreeding, depression in this respect was of lower extent in dioecious hemp than in monoecious hemp (Table 5). The seed yield of monoecious  $S_3$  plants was only 38.9% of that in the initial material, while in the dioecious F. Inb. this percentage was substantially higher compared to its own control. However, it must not be forgotten that in the  $S_0$  the thousand-seed-weight of the monoecious Fibrimon is much lower than that of the dioecious Kompolti, and this has a decisive influence on the seed yield of later generations. At the same time thousand-seed-weight is a surprisingly constant character in both dioecious and monoecious hems, showing a reduction of only 17% in the course of inbreeding. The order of the varieties is the same for seed yield and its components as for the other characters discussed.

(d) *Inbreeding coefficients.* Inbreeding coefficients were calculated for both the monoecious and dioecious varieties and strains, on the basis of the formula given in the Material and Method section.

In the case of monoecious and hermaphroditic species that can be self-pollinated, the trend for the inbreeding coefficient is well-known; here  $N_e = 1$ . In dioecious species which cannot be self-pollinated  $N_e = 2$  even in the case of the strictest sister line breeding. Accordingly, the F-coefficients in the successive generations show the trend seen in Table 6. Thus,

in  $S_8$  monoecious hemp practically reaches the absolute minimum, but the value of the inbreeding coefficient is noteworthy even in  $S_3$  and  $S_4$ , as confirmed by the data of the present experiments; dioecious hemp, on the other hand, has an inbreeding coefficient of 0.907 in  $S_{13}$ . The inbreeding coefficient of the dioecious varieties included in the experiment showed the following trend:

$$\begin{aligned} \text{F. Inb. } (S_{28}) &= 1 - (1 - 6)^{28} = 0.999 \\ \text{T. Inb. } (S_{13}) &= 1 - (1 - 6)^{13} = 0.907 \\ \text{S. Inb. } (S_{13}) &= 0.907 \end{aligned}$$

Besides the normal self-pollinated mother plants 4 sibbed  $S_2$  monoecious strains were available, for which inbreeding coefficients were also calculated depending on the size of the population. In this case  $S_1$  was a self-pollinated plant, i.e. one with an  $F$  value of 0.5, and there were different numbers of progenies.

Table 6  
*Inbreeding coefficients  
for monoecious and dioecious hems*

Inbred generation	Monoecious: $F$ $N_e = 1$	Dioecious: $F$ $N_e = 2$
$S_1$	0.500	0.167
$S_2$	0.750	0.306
$S_3$	0.875	0.422
$S_4$	0.937	0.519
$S_5$	0.969	0.599
$S_6$	0.984	0.666
$S_7$	0.992	0.722
$S_8$	0.997	0.768
$S_{13}$	0.998	0.907
$S_{28}$	0.999	0.999

On the basis of the formula  $F = 1 - \left(1 - \frac{1}{2}\right) \left(1 - \frac{1}{2N}\right)$  these showed the following trend:

strain No. $S_2$ -83	$F = 0.503$	98 plants
„ „ $S_2$ -85	$F = 0.504$	65 „
„ „ $S_2$ -87	$F = 0.518$	14 „
„ „ $S_2$ -88	$F = 0.504$	68 „

These figures give a numerical answer to the question of why fibre hemp stands derived from sibbing gave consistently higher values for yield and other characters than the generation obtained by inbreeding a single plant.

The inbreeding depression for the characters examined thus runs parallel with the increase in inbreeding coefficients. Depression shows a considerably faster rate in monoecious strains than in dioecious hems, because—as a marginal case of diploids—inbreeding carried out through the self-pollination of a single plant causes the  $F$  value to increase much faster. At the same time, in dioecious hems the inbreeding minimum (0.997) required for the limit of inbreeding attainable with 2 plants is not reached until  $S_{20}$ .

\*

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## References

- ALEXANDER, M. P. (1969): Differential staining of aborted and nonaborted pollen. Sugarcane Breeding Institute, Coimbatore 7-India, (Printed in USA), **44-3**, 111-122.
- BÓCSA, I. (1958): A kender beltenyésztésének újabb jelenségei (Recent phenomena in hemp inbreeding). *Növénytermelés*, **7**, 1-10.
- BÓCSA, I.—KISKÉRI, R.—BUGLOS, J. (1974): Die Wirkung der Inzucht auf die Fertilität des Luzernepollens. *Z. Pflanzenzüchtung*, **73**, 287-291.
- FLEISCHMANN, R. (1934): Der Einfluss der Tageslänge auf die Entwicklung von Hanf und Ramie. *Faserforsch.*, **11**, 156-161.
- FLEISCHMANN, R. (1941): Adatok a kender műveleti alakjának leírásához (Contribution to the description of the cultivated form of hemp). Thesis, Budapest.
- FRUWIRTH, C. (1922): Zur Hanfzüchtung. *Z. Pflanzenzüchtung*, **3**, 340-401.
- STEPANOV, G. S. (1975): The inbreeding method in hemp breeding. *Vesnik selskogospod nauki*, **5**, 58-61.
- SVÁB, J. (1971): A populációgenetika alapjai (Fundamental population genetics). *Mezőgazdasági Kiadó*, Budapest.

INHERITANCE OF FLESH COLOUR, SEED COAT CRACKS AND TOTAL  
SOLUBLE SOLIDS IN WATERMELON AND THEIR GENETIC RELATIONS  
II. QUANTITATIVE CHARACTERS AND THE ASSOCIATION  
BETWEEN VARIOUS CHARACTERS

It is important to know the genetic behaviour of total soluble solids in watermelon and the relationship between this character and others, especially flesh colour. Thus, this study was carried out in order to determine this behaviour and to examine the effect of genetic variance and environmental factors on soluble solids.

The mode of inheritance of this character was studied by FILOV—TOSCEV (1968) and SUZUKI—HALL (1971).

Six crosses were obtained from Congo, Kaho and Leeby cvs. and were used in this investigation in the period from 1972 to 1975 at the Faculty of Agriculture, Cairo University, Giza. The parents,  $F_1$ ,  $F_2$  and Bc generations were planted as mentioned in Part I of this study (ABD EL-HAFEZ *et al.* 1980).

The crosses were:

1. Kaho  $\times$  Leeby, 2. Leeby  $\times$  Kaho, 3. Congo  $\times$  Leeby, 4. Leeby  $\times$  Congo, 5. Kaho  $\times$  Congo, 6. Congo  $\times$  Kaho.

The quantitative character total soluble solids was measured with a hand refractometer. The linkage between qualitative characters and the association between qualitative and quantitative characters was studied.

To reveal the mode of inheritance of total soluble solids the following statistical techniques were used.

Dominance was calculated by:

1. comparing the expected arithmetic mean with the observed mean of the  $F_1$ ,  $F_2$  and  $BC_{P_2}$  generations (POWERS *et al.* 1950);
2. the relative potency of gene sets (WIGAN 1944, MATHER 1949);
3. Pearson's first and second coefficient of skewness.

The nature of gene action was determined by comparing the expected means of the  $F_1$ ,  $F_2$ ,  $BC_{P_2}$  and  $BC_{P_1}$  populations on the basis of arithmetic and geometric gene action with the observed mean of each population (POWERS *et al.* 1950, CHARLES—SMITH 1939).

The number of genes controlling the differences between the parents was estimated using the Castle-Wright (CASTLE 1921) and Wright formulae (BURTON 1951).

Heritability values measure the relative magnitude of genetic and non-genetic variance. Heritability in the broad sense refers to the ratio of heritable variance to total variance. Heritable variance includes the dominance, epistatic and additive genetic variances, while the total variance includes these plus environmental variance.

Heritability was calculated as follows:

$$H = \frac{V_{F_2} - V_E}{V_{F_2}} \times 100$$

where:  $V_{F_2}$  = variance of the  $F_2$

$V_E$  = environmental variance

Three estimates can be used for the environmental variance:

1.  $F_1$  variance ( $V_{F_1}$ ) (BURTON 1951).

2. The arithmetic mean of the two parental variances (FREY *et al.* 1954).

3. The cube root of the product of the variance of  $P_1$ ,  $P_2$  and  $F_1$  (WEBER—MOORTHY 1952, FIUZAT—ATKINS 1953).

In order to study the association between characters,  $F_2$  data of straight and reciprocal crosses were combined together and used to reveal linkage and association. Values of  $\chi^2$  were applied for testing these paired characters (HAYES *et al.* 1955).

#### Quantitative characters. Total soluble solids

This character was studied in the following crosses:

- |                        |                         |
|------------------------|-------------------------|
| 1. Kaho $\times$ Leeby | 3. Congo $\times$ Leeby |
| 2. Leeby $\times$ Kaho | 4. Leeby $\times$ Congo |

The study was not continued with crosses 5 and 6 because of the insignificant differences between the parents.

#### Crosses 1 and 2

The data are presented in Tables 1 and 2. The fruits of Kaho had an average total soluble solid content of  $7.41 \pm 1.09\%$ , whereas this percentage in Leeby was  $5.52 \pm 1.28\%$ . A difference of 1.89% in total soluble solids between the parents was significant.

Table 1

Comparative total soluble solids data for observed and theoretical means, mean differences, potency, skewness and minimum number of genes in the cross Kaho  $\times$  Leeby

Generation	P <sub>1</sub> Kaho	P <sub>2</sub> Leeby	F <sub>1</sub>	F <sub>2</sub>	BC <sub>P<sub>2</sub></sub>
No. of observed plants	16	22	13	72	21
Observed mean	7.41	5.52	5.95	6.17	5.82
Arithmetic mean			6.47	6.21	5.74
Geometric mean			6.40	6.16	5.83
Mean difference:					
Observed vs. observed	1.89*				
Observed vs. arithmetic			0.52 <sup>n.s.</sup>	0.04 <sup>n.s.</sup>	0.08 <sup>n.s.</sup>
Observed vs. geometric			0.45 <sup>n.s.</sup>	0.01 <sup>n.s.</sup>	0.01 <sup>n.s.</sup>
Potency			0.55	+0.32 <sup>n.s.</sup> + 0.65 <sup>n.s.</sup>	
Skewness					
CV%	14.72	23.17	14.72	20.22	22.28

Minimum number of genes (Castle—Wright formula) = 2.56

Minimum number of genes (Wright formula) = 3.17

Heritability % using first equ. = 11.19, using second equ. = 9.18, and using third equ. = 3.68

Table 2

*Comparative total soluble solids data for observed and theoretical means, mean differences, potency, skewness and minimum number of genes in the cross Leebby × Kaho*

Generation	P <sub>1</sub> Leeby	P <sub>2</sub> Kaho	F <sub>1</sub>	F <sub>2</sub>	BC <sub>P<sub>2</sub></sub>
No. of observed plants	22	16	19	81	39
Observed mean	5.52	7.41	6.51	6.16	6.78
Arithmetic mean			6.47	6.48	6.96
Geometric mean			6.40	6.27	7.09
Mean difference:					
Observed vs. observed		1.89*			
Observed vs. arithmetic			0.04 <sup>n.s.</sup>	0.32*	0.18 <sup>n.s.</sup>
Observed vs. geometric			0.11 <sup>n.s.</sup>	0.11 <sup>n.s.</sup>	0.31 <sup>n.s.</sup>
Potency			0.04		
Skewness				+0.21 <sup>n.s.</sup> + 0.40 <sup>n.s.</sup>	
CV%	23.17	14.72	19.22	22.86	19.32

Minimum number of genes (Castle-Wright formula) = 1.07

Minimum number of genes (Wright formula) = 0.11

Heritability % using first equ. = 21.01, using second equ. = 28.69, and using third equ. = 26.85

When comparing the observed means for the F<sub>1</sub> generations versus their arithmetic means, non-significant differences were obtained, indicating the absence of dominance. This trend was clear in both F<sub>2</sub> and BC<sub>P<sub>2</sub></sub>. The potency ratios were 0.25 and 0.04 respectively in the two crosses, which indicate the absence of dominance. The values of skewness for F<sub>2</sub> and BC<sub>P<sub>2</sub></sub> were insignificant. These data supported the absence of dominance.

The difference between the observed mean and the corresponding arithmetic and geometric means in F<sub>1</sub>, F<sub>2</sub> and BC<sub>P<sub>2</sub></sub> were practically insignificant in both parents. Thus, the nature of the gene action could not be revealed.

The minimum number of genes determined by the two formulae used was 2.56 and 3.17 respectively, i.e. three pairs of genes in the straight cross, and 1.07 and 0.11, i.e. one pair of genes in the reciprocal cross. The small number obtained using the Wright formula in the reciprocal cross might be due to the lack of certain assumptions necessary for a reliable estimate.

The heritability values ranged from 3.68 to 11.19 in the first cross and from 21.01 to 28.69 in the second cross, indicating that environmental conditions had a considerable effect on this character.

The coefficients of variability were 14.72, 23.17, 14.72, 20.22 and 22.86 for P<sub>1</sub>, P<sub>2</sub>, F<sub>1</sub>, F<sub>2</sub> and backcross respectively in the straight cross and 19.22, 22.86 and 19.32 for F<sub>1</sub>, F<sub>2</sub> and BC<sub>P<sub>2</sub></sub> respectively in the reciprocal cross.

#### Crosses 3 and 4

As shown in Tables 3 and 4, the means of the two parents were significantly different. The difference between the observed and the expected means for F<sub>1</sub> populations of both crosses was significant, indicating the presence of some sort of dominance. The F<sub>1</sub> means coincided with the value for the Leebby cultivar, indicating the complete dominance of the small percentage. This was confirmed by the potency ratio, which was 0.98 and 1.08 respectively for the two crosses. When comparing the observed means for F<sub>2</sub> and the first backcross generations versus their arithmetic means, significant differences were found for the F<sub>2</sub> of the straight cross and the BC<sub>P<sub>2</sub></sub> of the reciprocal cross, indicating partial and complete



Table 3

*Comparative total soluble solids data for observed and theoretical means, mean differences, potency, skewness and minimum number of genes in the cross Congo × Leeby*

Generation	P <sub>1</sub> Congo	P <sub>2</sub> Leeby	F <sub>1</sub>	F <sub>2</sub>	BCP <sub>2</sub>
No. of observed plants	12	35	13	65	30
Observed mean	7.13	5.46	5.48	6.31	5.55
Arithmetic mean			6.30	5.89	5.47
Geometric mean			6.24	5.85	5.87
Mean difference:					
Observed vs. observed	1.67*				
Observed vs. arithmetic			0.82*	0.42*	0.38 <sup>n.s.</sup>
Observed vs. geometric			0.76*	0.46*	0.32 <sup>n.s.</sup>
Potency			0.98		
Skewness				-0.90 <sup>n.s.</sup> — 0.84 <sup>n.s.</sup>	
CV%	15.89	19.01	15.18	17.69	20.57

Minimum number of genes (Castle—Wright formula) = 1.01

Minimum number of genes (Wright formula) = 2.98

Heritability % using first equ. = 44.45, using second equ. = 5.44, and using third equ. = 34.25

Table 4

*Comparative total soluble solids data for observed and theoretical means, mean differences, potency, skewness and minimum number of genes in the cross Leeby × Congo*

Generation	P <sub>1</sub> Leeby	P <sub>2</sub> Congo	F <sub>1</sub>	F <sub>2</sub>	BCP <sub>2</sub>
No. of observed plants	35	12	21	76	30
Observed mean	5.46	7.13	5.39	5.82	5.79
Arithmetic mean			6.30	5.84	6.26
Geometric mean			6.24	5.80	6.44
Mean difference:					
Observed vs. observed		1.67*			
Observed vs. arithmetic			0.91*	0.02 <sup>n.s.</sup>	0.47*
Observed vs. geometric			0.85*	0.02 <sup>n.s.</sup>	0.65*
Potency			1.08		
Skewness				+0.18 <sup>n.s.</sup> + 0.58 <sup>n.s.</sup>	
CV%	19.01	15.86	12.13	20.60	22.50

Minimum number of genes (Castle—Wright formula) = 0.44

Minimum number of genes (Wright formula) = 0.63

Heritability % using first equ. = 54.55, using second equ. = 18.05, and using third equ. = 43.40

dominance respectively for the small percentage. A non-significant skewness for  $F_1$  and  $F_2$  was found in these crosses.

When the observed  $F_1$ ,  $F_2$ ,  $BC_{P_1}$  and  $BC_{P_2}$  means were compared with the corresponding geometric means, no clear trend could be detected.

The minimum number of genes controlling the inheritance of this character as computed by the Castle—Wright and Wright formulae were 1.01 and 2.98 in the straight cross, and 0.44 and 0.63 in the reciprocal cross respectively, i.e. one to three pairs of genes. The small number of genes in the latter cross might be due to the lack of certain assumptions necessary for obtaining reliable figures.

When the heritability was computed using the first and third equations the estimates were fairly close to each other in both crosses (44.45 and 34.25 in the straight cross and 54.55 and 43.40 in the reciprocal cross), but the second equation gave a much lower estimate (5.44 and 18.05 in the two crosses respectively). This was due to the relatively large variances for this character in the parents. These heritability values indicate that environmental factors have some effect on this character.

The coefficient of variability ranged from 15.18 to 20.57% and from 12.13 to 22.50% in various populations in the straight and reciprocal cross respectively.

The studies were in accordance with those of SUZUKI—HALL (1971) who found in three crosses between Crimson Sweet and New Hampshire Midget that the total soluble solids was determined by three incompletely dominant genes  $su_1$ ,  $su_2$  and  $su_3$ . However, FILOV—TOSCEV (1968) found that of 88 hybrids, 27 excelled the better parent, 31 were intermediate, 11 resembled the seed parent, 12 the pollen parent and 7 were below both.

#### *Association between characters. Linkage*

In order to study linkage,  $F_2$  data derived from several crosses were used to test the independence of segregation in pairs of characters in all possible combinations, as presented in Table 5.

Table 5

$\chi^2$  values for testing independence of segregation of some characters in different watermelon crosses

Linkage				
Pairs of characters and crosses	N	Ratio	$\chi^2$	P
Flesh colour vs. cracks on seed coat:				
Kaho $\times$ Leeby	166	9 : 3 : 3 : 1	6.0590	0.10-0.90
Congo $\times$ Leeby	145	9 : 3 : 3 : 1	4.9202	0.20-0.50
Association between characters				
Characters associated	D.F.	$\chi^2$	P	
1. Flesh colour vs. total soluble solids:				
Kaho $\times$ Leeby	3	22.5786	0.01	
Congo $\times$ Leeby	3	25.9261	0.01	
2. Cracks on seed coat vs. total soluble solids:				
Kaho $\times$ Leeby	3	1.7039	0.30-0.50	
Congo $\times$ Leeby	3	0.5324	0.90-0.95	

The segregation in  $F_2$  populations between the two characters compared was tested for the ratio 9 : 3 : 3 : 1, which is found if each is governed by one pair of genes. There was a good fit to this ratio in all cases between flesh colour and cracks on seed coats.

*Association between qualitative and quantitative characters*

All possible combinations of qualitative and quantitative characters were tested for independence and the results of the chi-squared test of independence are shown in Table 5. No association was found between these two pairs of characters except between flesh colour and total soluble solids.

Data on the relationship between these two pairs of characters indicate that flesh colour was completely associated with the percentage of total soluble solids. In other words, in case of the cross Kaho  $\times$  Leeby, if the flesh was orange, the percentage of total soluble solids would be high, while the whitish-yellow flesh was associated with a low percentage. But in the case of the cross Congo  $\times$  Leeby, red flesh was associated with a high percentage whereas whitish-yellow flesh was associated with a low percentage. This complete association may be due either to the pleiotropy of the genes or to the fact that the loci of the genes responsible for these two characters are very close to each other and to the centromere (RILEY 1948). Moreover, these data explain why there was an absence of dominance in the  $F_1$  of Kaho  $\times$  Leeby, whereas the  $F_1$  of Congo  $\times$  Leeby showed complete dominance for the parent with a low percentage of total soluble solids. Nothing is reported in the available literature about this association in watermelon.

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### References

- ABD EL-HAFEZ, A. A.—GAAFER, A. K.—ALLAM, A. M. M. (1980): Inheritance of flesh colour, seed coat cracks and total soluble solids in watermelon and their genetic relations. I. Qualitative characters. *Acta Agron. Hung.* (in press).
- BURTON, G. W. (1951): Quantitative inheritance in pearl millet, *Pennisetum galucum*. *Agr. J.*, **43**, 409–417.
- CASTLE, W. E. (1921): An improved method of estimating the number of genetic factors concerned in cases of blending inheritance. *Science*, **54**, 223.
- CHARLES, D. R.—SMITH, H. (1939): Distinguishing between two types of gene action in quantitative inheritance. *Genetics*, **24**, 34–48.
- FILOV, A. T.—TOSCEV, V. P. (1968): Inheritance of sugar content in melons. *Cytologij i Genetika*, **2**, 144–151.
- FIUZAT, K. G.—ATKINS, R. B. (1953): Genetic and environmental variability in segregating barley populations. *Agron. J.*, **45**, 414–420.
- FREY, K. G.—SHEKLETON, M. C.—HALL, H. H.—BENNE, E. J. (1954): Inheritance of niacin, riboflavin and protein in two coat crosses. *Agron. J.*, **46**, 137–139.
- HAYES, H. K.—IMMER, F. R.—SMITH, D. C. (1955): *Methods of Plant Breeding*. McGraw-Hill Publishing Company Ltd. New York—London—Toronto, 551.
- MATHER, K. (1949): *Biometrical Genetics*. Dover Publications, Inc., 158.
- POWERS, L.—LOCKE, F.—GARRETT, J. C. (1950): Partitioning method of genetic analysis applied to quantitative characters of tomato crosses. *U.S.D.A. Tech. Bull.*, 998.
- RILEY, H. P. (1948): *Introduction to Genetics and Cytogenetics*. John Wiley and Sons, Inc., New York, 596.
- SUZUKI, Y.—HALL, C. V. (1971): Inheritance of melon weight, total soluble solids, seed size and number, and rind thickness and toughness in watermelon. *Hort. Science*, **6/3** sect. 2.
- WEBER, C. R.—MOORTHY, B. R. (1952): Heritable and nonheritable relationship and variability of oil content and agronomic characters in the  $F_2$  generation of soy bean crosses. *Agron. J.*, **44**, 202–209.
- WIGAN, L. G. (1944): Balance and potency in natural populations. *J. Genetics*, **46**, 150–160.



EFFECT OF WATER STRESS ON PEROXIDASE ACTIVITY  
AND ISOZYMIC PATTERNS IN TWO *CICER ARIETINUM* L. CULTIVARS  
WITH DIFFERING SENSITIVITIES TO WATER STRESS

In an earlier study on *Cicer arietinum* cv. C 214 and G 130 in relation to water stress, cv. C 214 was adjudged to be relatively drought resistant and cv. G 130 susceptible during early vegetative growth (SINGH—RAI 1980). The higher proline accumulation and drought resistance relationship proposed by SINGH *et al.* (1972) was found to be unreliable in *Cicer arietinum* when tested with the above cultivars (SINGH—RAI 1981). MALI—MEHTA (1977a, b) studied peroxidase activity and its electrophoretic patterns in rice cultivars with different sensitivities to water stress, and suggested that peroxidases could provide information on the drought resistance phenomenon of the plants. Such comparative studies are lacking for legumes. It is our contention that a comparison of two differently sensitive cultivars of the same crop can give an insight into the phenomenon of drought resistance.

Seedlings of *Cicer arietinum* L. (cultivars C 214, resistant, and G 130, susceptible) were grown in a water culture and were subjected to different levels of water stress (from 0 to -10 atm by dissolving PEG-6000) at the age of 10 days. Studies on isozymic patterns and anzymic assays were conducted on the eighth day under water stress. WSD was measured after WEATHERLEY—SLATYER (1957).

*Electrophoretic pattern of peroxidase*

The tissues (leaves and roots) were homogenized in 0.05 M Tris-HCl buffer at pH 7.8<sup>9</sup> and centrifuged at 12 000 rpm at 0.4 °C for 20 minutes. The supernatant was dialysed against distilled water at 5 °C. The dialysed enzyme solution was used for an electrophoretic run. The procedure followed for isozymic studies was essentially that of DAVIES (1964) using alkaline buffers. The sample gel and spacer gel were, however, omitted and a sample mixed with 60% sucrose (1 : 2) was layered directly on the top of a running gel of 7.5% strength. A constant current of 4 mA/tube was applied and the electrophoresis was accomplished in 50–60 min, when the bromophenol blue dye travelled up to 0.5 cm from the bottom. After the run the gels were squeezed out with a jet of water and stained for peroxidase in a saturated solution of benzidine and 1% H<sub>2</sub>O<sub>2</sub> for one minute, after which blue bands appeared.

*Peroxidase assay*

Peroxidase activity was determined after SRIVASTAVA—VAN HUYSTEE (1973). An enzyme aliquot was prepared by homogenizing 250 mg of different seedling parts (leaves and roots) in 5 ml of chilled 0.2 M sodium phosphate buffer (pH 6.0) and centrifuging at 5000 g at 0 °C for 11 min. The rate of decomposition of H<sub>2</sub>O<sub>2</sub> by the enzyme, with O-diansidine as the hydrogen donor, was determined spectrophotometrically by measuring the colour development at 460 nm. Enzyme activities were calculated both per unit fresh weight and per unit protein.

*Electrophoretic patterns of peroxidase*

The isozymic electrophoretic pattern of peroxidase in unstressed and stressed seedlings of resistant cv. C 214 and susceptible cv. G 130 in shoot and root on the 8th day of stress is shown in Fig. 1. The shoots of both cultivars show only two bands (R<sub>m</sub> 0.18 and 0.39). With an increase in stress levels in cv. C 214 band No. I became both lighter and thinner beyond

—6 atm, while the intensity of band II increased beyond —3 atm. In cv. G 130 band I became lighter only at —10 atm, while band II regularly increased its intensity up to —10 atm.

The roots exhibited characteristic differences between the cultivars. Control plants of cv. C 214 have only three isozymic bands (Rm 0.14, 0.36 and 0.53) the intensity of which did not change with increasing stress levels, though the width definitely increased. As a result of

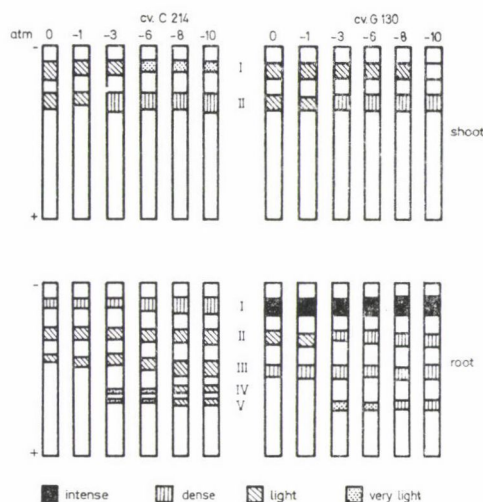


Fig. 1. Zymogram showing the electrophoretic pattern of peroxidase at different levels of water stress in the shoots and roots of cv. C 214 and cv. G 130 on the 8th day of stress

stress two new bands (IV, V) appeared at —3 atm and increased both in intensity and width up to —10 atm. In the roots of cv. G 130, band I was very prominent and intense, while band II was light in the controls but became dense beyond —3 atm. Band IV was missing in the roots of cv. G 130, while band V appeared at —3 atm and increased in intensity and width up to —10 atm. Thus, cultivar differences were more clearly expressed in the roots than in the shoots.

In the shoots and roots of both the cultivars the total peroxidase activity expressed on a per gram fresh weight basis increased with an increasing level of water stress (Table 1). A significant increase in the activity was observed even at —1 atm except in the roots of cv. C 214, where it showed a significant increase at —3 atm (Table 1). Both the actual values and the percentage increase over the control of peroxidase activity were significantly higher in the susceptible cv. G 130 than in the resistant cv. C 214 in both the shoots and the roots (Table 1).

However, the peroxidase activity expressed on a per mg protein basis presented an entirely different picture and a decrease in activity was clearly shown as the stress levels increased (Table 1 and Fig. 2). A clearcut cultivar response was shown, i.e. a significantly higher peroxidase activity was shown in the shoots and the roots in the resistant cv. C 214. However, the percentage inhibition of peroxidase activity was higher in the resistant cv. C 214 than in cv. G 130 in the case of the shoots, while the roots presented no specific trend (Fig. 2). There exists a negative correlation (shoot:  $r = -0.6951$ , root:  $r = -0.611$ ) between peroxidase activity and the WSD changes (Fig. 2).

Table 1

*Peroxidase activity in the shoots and roots of cv. C 214 and cv. G 130 of Cicer arietinum at different levels of water stress after 8 days of stress*

		Peroxidase activity							
		$\Delta$ OD at 460 nm/g fresh wt./min.				$\Delta$ OD at 460 nm/mg protein/min.			
		Shoot		Root		Shoot		Root	
		cv. C 214	cv. G 130	cv. C 214	cv. G 130	cv. C 214	cv. G 130	cv. C 214	cv. G 130
atm	0	121.0	139.5	149.5	140.0	18.7	9.81	42.15	30.87
	-1	122.5	141.0	149.0	149.0	17.1	9.27	37.64	25.02
	-3	128.3	146.0	162.5	155.0	9.68	6.59	32.49	28.53
	-6	129.0	159.0	155.0	154.5	9.20	5.36	31.56	22.05
	-8	136.0	160.5	156.5	157.0	8.60	4.42	17.99	12.97
	-10	138.0	166.0	158.0	158.0	7.52	5.04	15.90	14.22

		Fresh weight basis		Per mg protein basis	
C.D. (0.05)	To compare varieties	Shoot	0.75	2.80	
		Root	1.10	4.60	
	To compare stress	Shoot	1.30	4.80	
		Root	1.91	3.01	

In the present studies the isozymic patterns of peroxidase in *Cicer arietinum* L. cv. C 214 and cv. G 130 are shown to be altered in relation to water stress. In the shoots this effect was only manifested by changes in the intensity or breadth of the individual isozymic bands. The first isozymic band showed more stability in the susceptible cv. G 130, where a decrease in intensity was only shown at -10 atm, as compared to the resistant cv. C 214, where the intensity was reduced even at -6 atm. Changes were also recorded in the second isozymic band, and higher intensity was a characteristic of the susceptible cv. G 130. The two cultivars showed characteristic differences with regard to root peroxidases, and the first and third bands were of higher intensity in the susceptible cv. G 130 than in cv. C 214. The characteristic cultivar differences were marked by the appearance of two new isozymic bands in the roots of cv. C 214 at Rm 0.77 (IV) and 0.83 (V), and of band V only in the roots of cv. G 130. MALI—MEHTA (1977a) observed a decrease in the intensities of individual isozymic bands in stressed seedlings of rice cultivars. STUTTE—TODD (1969) reported the disappearance of certain peroxidase isozymic bands and the appearance of new ones in relation to water stress in wheat.

The increase in shoot/root peroxidase activity (on a per g fresh weight basis) in cv. C 214 is clearly supported by an increase in the number and/or intensity of the isozymic bands. The significantly higher peroxidase activity (on a fresh weight basis) in both the shoots and roots of cv. G 130 is also reflected in the higher band intensity of cv. G 130 isozymic bands.

Peroxidase activity (per mg protein) declined with increasing levels of water stress in *Cicer arietinum*. A decline in the peroxidase activity under water stress has been recorded by LUKICHEVA (1968) in spring wheat and MALI—MEHTA (1977b) in the TKM-I and I.S. cultivars of rice after 72 hr of inhibition. No definite relationship between peroxidase activity and changed water stress could be observed by TAKESHI (1968) in the leaves of pea or MALI—MEHTA (1977b) in the shoots of cv. I.S. of rice. However, STUTTE—TODD (1969) and MALI—



MEHTA (1977b) reported increased peroxidase activity (per mg protein) with increased stress in wheat and in the roots of rice cv. TKM-I.

WSD increased with water stress in both the cultivars, the extent of increase being higher in the susceptible cultivar G 130 (Fig. 2). WSD was negatively correlated with the peroxidase activity in *Cicer arietinum* ( $r = -0.695$  shoot,  $-0.611$  root). Growth in terms of

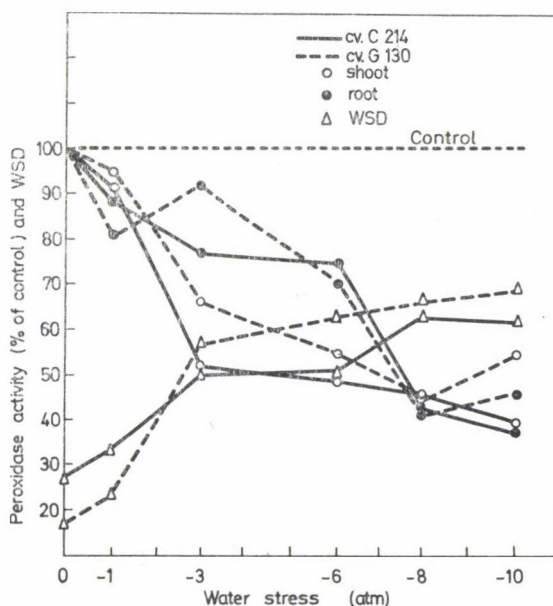


Fig. 2. The effect of water stress on the peroxidase activity of cv. C 214 (—) and cv. G 130 (---) in shoots (○) and roots (●) and WSD in cv. C 214 (—) and cv. G 130 (---)

dry weight was found to decrease in *Cicer arietinum* L. with an increasing level of water stress, the susceptible cultivar exhibiting a higher degree of growth inhibition. In *Cicer arietinum*, peroxidase activity and growth showed a positive correlation under water stress.

The two cultivars behaved in a different manner, with higher total peroxidase activity being shown in the resistant cv. C 214 than in the susceptible cv. G 130. However, a higher percentage inhibition of peroxidase activity was shown in the shoots of the resistant cultivar and no specific pattern could be shown in the roots of the two cultivars.

Many of the water stress effects have been shown to be due to increased levels of abscisic acid (ABA) under stressed conditions (WRIGHT—HIRON 1969, MILBORROW 1974) and ABA has also been shown to affect peroxidase activity. MEGHA—LALORAYA (1977) showed that in *Trigonella* seedling axis. Although ABA inhibited growth in terms of fresh weight, it also promoted peroxidase activity (per g fresh weight basis). Similarly etheral or gallic acid, which also inhibited growth in *Trigonella*, promoted peroxidase activity (per fresh weight basis). Thus, there seems to be a clear-cut correlation between growth inhibition and peroxidase activity (MEGHA—LALORAYA 1978a, b). In the present set of experiments, although water stress inhibits growth both in terms of fresh and dry weights (SINGH—RAI 1980) there is a promotion in peroxidase activity (on a fresh weight basis). However, whether this is a

direct effect of water stress or a secondary effect manifested by changed ABA levels under water stress is not clear.

The drought resistant cv. C 214 of *Cicer arietinum* is characterized by higher peroxidase activity at all levels of water stress. Similar results were reported by MALI—MEHTA (1977b), where higher peroxidase activity was shown to be characteristic of the drought tolerant cv. TKM-I of rice at the 72 hr inhibition stage. However, there is a need for more data to accumulate on peroxidase enzymic behaviour towards water stress in the resistant and susceptible cultivars of different crops before any definite role in drought resistance can be proposed for peroxidase activity, as the resistant cultivar showed a higher percentage inhibition peroxidase activity in the shoots than the susceptible cv. G 130.

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#### References

- DAVIES, B. J. (1964): N. Y. Acad. of Sci. Ann. **121**, 404–427.  
 LUKICHEVA, E. L. (1968): Tr. Inst. Bot. Akad. Nauk Kaz. SSR. **25**, 23–29.  
 MALI, P. C.—MEHTA, S. L. (1977a): Phytochem. (Oxf.), **16**, 643–646.  
 MALI, P. C.—MEHTA, S. L. (1977b): Phytochem. (Oxf.), **16**, 1355–1358.  
 MEGHA, B. M.—LALORAYA, M. M. (1977): Biochem. Physiol. Pflanzen., **171**, 269–277.  
 MEGHA, B. M.—LALORAYA, M. M. (1978a): Biochem. Physiol. Pflanzen., **172**, 305–310.  
 MEGHA, B. M.—LALORAYA, M. M. (1978b): Biochem. Physiol. Pflanzen., **173**, 229–237.  
 MILBORROW, B. V. (1974): Ann. Rev. Pl. Physiol., **25**, 259–307.  
 SINGH, G.—RAI, V. K. (1980): Indian J. Ecol., **7**, 246–253.  
 SINGH, G.—RAI, V. K. (1981): Biologia Plant., **23**, 86–90.  
 SINGH, T. N.—ASPINALL, D.—PALEG, L. G. (1972): Nature New Biol., **236**, 188–190.  
 SRIVASTAVA, O. P.—VAN HUYSSTEE, R. B. (1973): Can. J. Bot., **51**, 2207–2215.  
 STUTTE, C. A.—TODD, G. W. (1969): Crop. Sci., **9**, 510–512.  
 TAKESHI, T. (1968): Bot. Mag. (Tokyo), **81**, 297–309.  
 WRIGHT, S. T. C.—HIRON, R. W. P. (1969): Nature, **224**, 719–720.  
 WEATHERLEY, P. E.—SLATYER, R. O. (1957): Nature, **179**, 1085–1086.

### INFLUENCE OF SOIL TYPES, PRE-SOWING TREATMENTS, AVAILABLE AMOUNTS OF SOIL NUTRIENTS AND THEIR COMBINATIONS ON TOMATOES.

#### III. EFFECT ON SEEDLING GROWTH AND FLOWERING

Flowering (ANGELOV 1974) and plant growth (FERENCZ *et al.* 1964) were affected by the differential application of N, P and K to the plant. Moreover, Uzo (1971) emphasized that not only are adequate amounts of N, P and K important for optimum tomato yield, but also a satisfactory balance between the amounts of the three nutrients. This depends on soil type. Pre-sowing seed treatments also have an effect on the growth (WOODRUFF 1969) and flowering of the plant (BESKROVNAYA 1970).



No information on the interaction effect of soil type, seed treatments and proportions of essential macro-nutrient elements available in the soil on the growth and flowering of tomato plants, was obtained from the literature. Thus, the present study was planned to determine to what extent the growth and (flower) anthesis of the plant can be influenced by the factors mentioned to figure as index of earliness and production capacity.

Full details of the treatments applied, the soil preparation with available nutrients and the cultural methods used have been reported in a previous paper (EL-SAWAH 1981).

Shallow wooden boxes ( $40 \times 60 \times 10$  cm), replicated 3 times, were filled with the soils under study. Each box contained 12 rows divided into 3 sets of 4 rows containing 104 seeds for each seed treatment, with 26 seeds in each row. When emergence was completed, the plants were thinned to 13 plants per row. On the 40th day 72 plants (treatment) representing 3 replicates were sampled and immediately transported to the laboratory for recording the following observations on seedling performance:

a) Stem length from the root neck to the first leaf of the plant (I) and to the growing tip (II).

b) Stem diameter.

c) Fresh and dry weight per plant (aerial parts) and dry weight per root.

The remaining seedlings in the boxes were left till most plants had flowered (75 days after sowing) and the opened flowers, abscised flowers, set flowers and total flowering were recorded as a percentage of the seedlings.

The results were analysed by standard analysis of variance with the differences between the means tested by Duncan's multiple range test. The signs \*, \*\*, \*\*\* and n.s. within the columns of the tables indicate results significantly different at the 5, 1, 0.1% levels and not significantly different, respectively. Also, values which are indicated with similar alphabetical letters do not differ significantly.

*Effect of available quantities and ratios of nutrients.* Concerning the dry and fresh weights of seedlings and the diameter and length of the stem, the data indicated that various quantities and ratios of N,  $P_2O_5$  and  $K_2O$  gave the same development and growth of seedlings (Table 1). There are three possible interpretations of this phenomenon (STEINER 1966).

I. A plant takes up ions in a mutual ratio, controlled by the mutual ratio of the ions in the soil solution, but the mutual ratio of the ions in the plant does not have a strong effect on development and growth.

II. The plant has a strong selective capacity, in other words a plant will absorb ions in its own mutual ratio, within wide limits, independently from the mutual ratio between the ions in the nutrient solution.

III. The effect of the quantities and ratios of available soil nutrients on plant growth depends on the growth phase, which in the present investigation was too early to show an effect on growth performance.

Regarding the flowering, if two nutrient element ratios were kept constant the increase in the quantity of the third nutrient element ratio resulted in an increase in the percentage of opened flowers, set flowers and total flowers. This was true with all treatments except when comparing the treatments 1 : 2 : 2, 2 : 2 : 1 and 2 : 1 : 2, and in the treatment 2 : 2 : 2, which caused a significant decrease in all the flowering features studied. In this connection, it was probable that K had no restricting effect, as shown by the slight difference between treatments 1 : 1 : 1 and 1 : 1 : 2, 1 : 2 : 1 and 1 : 2 : 2 and 2 : 1 : 2. Surplus N seemed to hinder flowering and flower setting. It was also interesting to compare treatments 1 : 1 : 1 and 2 : 2 : 2; the NPK ratios were similar, but they differed in absolute quantity. It is probable that the 1 : 1 : 1 ratio was not enough to satisfy the nutrient requirements, as shown by plant analysis for nutrient content and uptake (EL-SAWAH 1982), so it was insufficient for the production of enough flowers. On the other hand, although the 2 : 2 : 2 treatment was favour-



**Table 1**  
*Effect of nutrient ratio (N) and its interaction with soil type (T) on seedlings' growth and flowering*

Soil type	Nutrient ratio	Fresh Wt./plant, gm	Dry Wt./root	Dry Wt./plant	Stem			Flowering			
					length		diameter, mm	abscission, %	set, %	opened, fl. %	total, fl. %
					I.	II.					
				mg	cm						
Sand	1 : 1 : 1	3.843 abed	100	363 abc	5.06	13.9	4.03	16.6	21.9 b	37.7 cd	76.2 b
	1 : 1 : 2	4.144 abcde	106	430 abc	4.47	12.7	4.66	36.9	34.7 e	36.8 ed	108.4 ed
	1 : 2 : 1	4.839 cde	113	401 abc	4.80	14.0	4.38	23.2	25.7 bc	48.0 de	96.8 c
	1 : 2 : 2	5.062 cde	120	420 abc	5.27	15.3	4.49	28.1	35.5 c	47.1 de	110.7 cd
	2 : 1 : 1	4.779 bcde	109	434 bc	5.18	14.0	4.37	52.0	29.4 bc	68.7 e	150.2 e
	2 : 1 : 2	5.451 de	118	480 c	5.63	16.2	4.68	52.7	35.4 c	53.0 de	141.1 de
	2 : 2 : 1	5.573 e	112	481 c	5.33	15.3	4.75	43.6	31.1 bc	63.4 e	137.7 cde
	2 : 2 : 2	3.665 abc	105	348 abc	4.84	12.7	4.11	5.9	5.4 a	28.1 bc	39.3 ab
Loamy sand	1 : 1 : 1	3.691 abc	63	364 abc	4.80	11.2	3.61	7.1	1.9 a	15.7 ab	24.7 a
	1 : 1 : 2	2.784 a	64	294 a	4.35	9.9	3.58	3.8	0.5 a	6.2 a	10.5 a
	1 : 2 : 1	3.577 abc	69	339 ab	5.02	12.4	4.07	6.8	0.9 a	26.1 bc	33.8 a
	1 : 2 : 2	3.127 ab	72	305 ab	4.13	10.7	3.87	5.3	3.2 a	13.4 ab	22.0 a
	2 : 1 : 1	3.147 abc	58	330 ab	4.27	9.9	3.78	0.0	0.0 a	3.1 a	3.1 a
	2 : 1 : 2	3.701 abc	61	333 ab	4.70	11.6	3.96	2.6	2.5 a	8.8 a	14.0 a
	2 : 2 : 1	2.834 a	47	272 a	4.34	10.2	3.72	0.5	0.0 a	3.9 a	4.0 a
	2 : 2 : 2	3.650 abc	55	328 ab	4.87	11.9	3.90	3.1	0.9 a	8.2 a	12.3 a
N		n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	*	n.s.	*
N × T		*	n.s.	*	n.s.	n.s.	n.s.	n.s.	*	*	*

able for increasing both uptake and content of nutrients it has a causal effect, unfavourable for flowering, due probably to its influence on nutrient distribution within the plant tissue and on the C/N ratio. This was further attributed to the synthesis of protein substances at the expense of carbohydrate. From this discussion and from the non-statistical differences between other treatments regarding flowering it can be concluded that the stimulation of flowering was caused only by NPK supplies with different ratios, whereas those with equal NPK ratios significantly depressed flower production and also flower setting.

*Effect of soil type.* Table 2 shows that loamy sand soil had a depressive effect on the growth of both root and plant, which might be due to:

I. Size of pores: The failure of root penetration in compacted soils may be due to pore size rather than to lack of oxygen (VEIHMEYER—HENDRICKSON 1948);

II. Aeration: Diminuation of the elongation and development of both roots and growth are highly correlated with the oxygen diffusion rate (BERTRAND—KOHNE 1957) and aeration (TROUSE—BAVER 1962);

III. Bulk density: Elongation of roots in the soils was correlated with bulk density at 100 cm soil moisture tension, since aeration was non-limiting at this moisture content; the growth was correlated with both aeration and bulk density at 10 cm tension (PHILLIPS—KIRKHAM 1962); and/or

IV. Soil-moisture tension: Experiments on the penetration of roots into compressed cores where aeration was non-limiting demonstrated that root elongation at a given bulk density depended upon the soil moisture tension (TAYLOR—HERBERT 1963). The higher the moisture tension, the lower was the percentage penetration into the cores.

Concerning the flowering of plants as affected by soil types, the data also indicated that seedlings on sand had a markedly higher flower production when compared to loamy sand seedlings. This might be attributed to the high P content of sand seedlings.

*Effect of presowing treatments.* Table 2 indicates that pre-sowing treatment, either by soaking seeds in pure water or in nutrient solution, significantly increased both fresh and dry weights of seedlings as well as stem length and flowering when compared to the control. Moreover, although dry weight of roots and stem diameter had the same trend as the other characters studied, pre-sowing treatments did not reflect any significant differences between them. Irrespective of the dry weight of seedlings, there were no significant differences between seeds soaked in water and seeds soaked in Volldünger. WOODRUFF (1969) mentioned that due to thicker leaves, with thicker cell walls and more bound water, the dry weight increased in seedlings produced from pre-treated seeds compared to the untreated control. GAAROV (1971) also reported that plants produced from pre-treated seeds had decreased transpiration and cell sap concentration compared with plants from untreated controls.

With respect to flowering, the same results were obtained by BESKROVNAYA (1970). This good effect was perhaps due to the stimulation of translocation of metabolites towards the reproductive organs (ANISIMOV—NUSKOVA 1973, BELOKOBYL'SKÜ—MALYKIRA 1970, VLAYYUK *et al.* 1974).

*Effect of the combination of soil types with their nutrient quantity and ratios.* As presented in Table 1, it is clear that combined treatments are sometimes superior to single ones; this is true for the combination effects of soil types with the nutrients in the soil, which had a significant effect only on the fresh and dry weights of seedlings and on flowering. Thus in sandy soil larger seedlings are produced with 2 : 2 : 1, which only differed significantly with 1 : 1 : 1 and 2 : 2 : 2. Moreover, although no differences were obtained among the treatments in loamy sand, the treatment 1 : 2 : 1 seemed to be superior. This might be explained by the results obtained, which reflect the influence of the treatments indicated on stem length and diameter.

It should be mentioned that samples from the various treatments were taken at the

Table 2

*Effect of seed treatments, soil types and their interaction on seedlings' growth and flowering*

Soil type	Seed treatment	Fresh Wt./plant, gm	Dry Wt./ root	Dry Wt./plant mg	Stem			Flowering			
					length		diameter, mm	abscission, %	set, %	opened, fl. %	total, fl. %
					I.	II.					
					cm						
Sand	Dry seeds	4.61	106	404	4.93	13.3 c	4.43	27.5	25.9	45.5	98.8
	Seeds soaked in water	4.76	107	421	5.18	14.8 d	4.45	35.0	25.2	51.3	111.4
	Seeds soaked in Volldünger	4.64	117	434	5.11	14.7 d	4.42	34.8	31.0	46.7	112.4
	Mean of sand	4.67	110	420	5.07	14.2	4.43	32.0	27.4	47.8	107.2
Loamy sand	Dry seeds	3.21	59	315	4.60	10.6 a	3.73	3.8	1.9	9.2	15.0
	Seeds soaked in water	3.35	62	320	4.46	11.0 ab	3.82	4.9	0.3	10.6	15.8
	Seeds soaked in Volldünger	3.38	63	326	4.63	11.2 b	3.88	2.3	1.5	12.1	15.9
	Mean of loamy sand	3.31	61	321	4.57	11.0	3.81	3.7	1.2	10.6	15.6
Mean of seed treatments											
	Dry seeds	3.91 a	83	360 a	4.60 a	12.0 a	4.08	15.1	13.9 a	27.3 a	56.9 a
	Seed soaked in water	4.05 b	85	371 b	4.90 b	12.9 b	4.13	19.9	12.8 a	31.0 b	63.6 b
	Seeds soaked in Volldünger	4.01 b	90	380 c	4.87 b	12.9 b	4.15	18.5	16.2 b	29.4 b	67.2 b
Soil type (T)		***	***	***	*	***	***	***	***	***	***
Seed treatment (S)		*	n.s.	*	*	***	n.s.	n.s.	*	*	*
T×S		n.s.	n.s.	n.s.	n.s.	*	n.s.	n.s.	n.s.	n.s.	n.s.



same time for each treatment. It is clear, however, that after 40 days, for example, plants growing in sandy soil are more highly developed; in other words, they may be in another growth phase. So the differences found in fresh weight per plant may be due to the difference in growth phase, i.e. to the different level of development, which has a serious influence on the mutual ratio of uptake, which reflects the influence of nutrients on the growth.

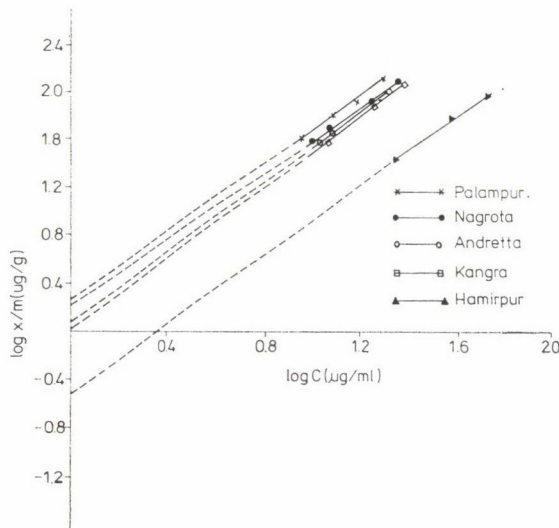


Fig. 1. Relationships between total phosphorus uptake per tomato seedling and % P content in plant top, and flowering

Concerning flowering, correlation coefficients and linear regressions were calculated for total P uptake per seedling and the % P content in plant tops, on the one hand, and total flowers %, fruit set % and opened flowers % on the other. These showed positive correlations (PROHÁSZKA—HAMAR 1977) and fitted a curvilinear regression fairly closely, as shown in Fig. 1, which also gives "r" values for the regression equations. However, the linear regressions show sufficient relationships to serve as illustration.

So the interaction effects of soil types and nutrient ratios were statistically affected only by P levels (EL-SAWAH 1982) and flowering; the discussion will be restricted to these factors.

Similar trends to those obtained were observed in the behaviour of plant flowering. The percentage of opened, total, set and abscised flowers on plants from sandy soil increased with the increasing P or K ratios in the soil when the N ratio was maintained at a ratio of 1, but when it was changed to 2 and increase in either of their ratios resulted in a decrease in all flowering indices. Moreover, except for the 2 : 2 : 2 treatment, which significantly reduced all flowering indices when compared with the treatments 1 : 2 : 2, 2 : 2 : 1 or 2 : 1 : 2, all flowering indices increased with an increase in the N ratio. On the other hand, a significant increase in total flower % was obtained with the treatments 2 : 1 : 1, 1 : 1 : 2 and 1 : 2 : 1, in opened flower % with 2 : 1 : 1 and in flower set % with 1 : 1 : 2 when compared with the treatment 1 : 1 : 1. The results also showed several significantly favourable combinations for flowering in ascending order of 2 : 2 : 1, 2 : 1 : 2 and 2 : 1 : 1. This can be partly explained

**Table 3**  
*Effect of seed treatments (S) × nutrient ratio (N) and their*

Soil type	Seed treatment	Nutrient ratio	Fresh Wt./plant, gm	Dry Wt./root, mg	Dry Wt./plant, mg	Stem
						length I. cm
Sand	Dry seed	1 : 1 : 1	3.44 abcdefgh	93	347	5.0 cdefghi
		1 : 1 : 2	3.79 abcdefghij	113	403	4.1 ab
		1 : 2 : 1	4.98 jklmno	105	335	4.6 abcdefg
		1 : 2 : 2	4.60 ghijklmno	106	416	5.1 defghij
		2 : 1 : 1	5.11 klmno	116	455	5.4 ghij
		2 : 1 : 2	5.73 no	107	505	5.9 j
		2 : 2 : 1	5.64 no	118	444	4.7 abcdefgh
		2 : 2 : 2	3.59 abcdefgh	92	331	4.6 abcdefg
	Seeds soaked in water	1 : 1 : 1	4.18 defghijkl	100	376	5.2 efghij
		1 : 1 : 2	4.76 hijklmno	105	436	4.6 abcdefg
		1 : 2 : 1	4.48 fghijklmn	112	411	5.0 cdefghi
		1 : 2 : 2	5.48 mno	129	424	5.2 efghij
		2 : 1 : 1	4.92 ijklmno	105	454	5.4 ghij
		2 : 1 : 2	5.25 lmno	112	466	5.6 ij
		2 : 2 : 1	5.31 lmno	104	469	5.9 j
		2 : 2 : 2	3.11 abcde	94	330	4.6 abcdefg
	Seeds soaked in Volldünger	1 : 1 : 1	3.91 bcdefghijk	107	365	4.9 bcdefghi
		1 : 1 : 2	3.88 abcdefghijk	101	451	4.7 abcdefgh
		1 : 2 : 1	5.06 klmno	123	458	4.8 abcdefghi
		1 : 2 : 2	5.11 klmno	124	419	5.5 hij
		2 : 1 : 1	4.31 efghijklm	108	396	4.8 abcdefghi
		2 : 1 : 2	5.37 lmno	132	470	5.4 ghij
		2 : 2 : 1	5.77 o	115	531	5.5 hij
		2 : 2 : 2	3.70 abcdefghi	130	384	5.3 fghij
Loamy sand	Dry seed	1 : 1 : 1	3.32 abcdef	69	328	4.9 bcdefghi
		1 : 1 : 2	2.67 ab	65	284	4.3 abcd
		1 : 2 : 1	3.36 abcdefg	82	326	5.0 cdefghi
		1 : 2 : 2	3.46 abcdefgh	63	343	4.2 abc
		2 : 1 : 1	3.10 abcde	55	306	4.2 abc
		2 : 1 : 2	3.93 cdefghijk	54	364	4.5 abcdef
		2 : 2 : 1	2.64 a	41	277	4.2 a
		2 : 2 : 2	3.24 abcdef	47	296	4.4 abcde
	Seeds soaked in water	1 : 1 : 1	4.20 defghijkl	60	417	4.6 abdeefg
		1 : 1 : 2	2.96 abcd	62	301	4.8 abcdefghi
		1 : 2 : 1	3.93 cdefghijk	64	365	4.9 bcdefghi
		1 : 2 : 2	2.82 abc	82	263	4.3 abcd
		2 : 1 : 1	2.87 abc	52	335	4.4 abcde
		2 : 1 : 2	3.50 abcdefgh	73	317	4.8 abcdefghi
		2 : 2 : 1	2.99 abcd	57	270	4.1 ab
		2 : 2 : 2	3.53 abdeefgh	47	296	5.1 defghij
	Seeds soaked in Volldünger	1 : 1 : 1	3.57 abcdefgh	61	347	4.9 bcdefghi
		1 : 1 : 2	2.72 abc	66	296	4.0 a
		1 : 2 : 1	3.45 abcdefgh	62	326	5.2 efghij
		1 : 2 : 2	3.10 abcde	72	310	4.1 ab
		2 : 1 : 1	3.47 abcdefgh	68	348	4.2 abc
		2 : 1 : 2	3.88 abcdefghijk	57	335	4.3 abcd
		2 : 2 : 1	2.88 abc	43	272	4.8 abcdefghi
		2 : 2 : 2	4.18 defghijkl	72	378	5.1 defghij
	S × N		n.s.	n.s.	n.s.	n.s.
	T × S × N		*	n.s.	n.s.	*

interaction with soil types (T) on seedling growth and flowering

length II.	diameter, mm	abscis- sion, %	set, %	Flowering	
				opened, fl. %	total, fl. %
13.4 jklmno	4.02 bcdefghi	10.3	22.8 defg	39.4 cdefghijklm	72.5 defg
10.9 bcdefg	4.59 fghijkl	39.8	28.5 fghij	36.0 bcdefghijklm	104.2 fgh
12.9 hijklmn	4.18 cdefghi	20.0	22.1 defg	43.4 efghijklm	85.5 fgh
13.3 ijklmno	4.26 cdefghijk	27.5	36.9 ghijk	42.3 cdefghijklmn	106.6 fghi
13.8 klmnop	4.66 hijkl	52.4	26.2 fghi	59.3 lmn	137.9 hijk
16.0 rst	4.93 kl	44.0	47.7 k	62.5 mno	154.3 ijkl
14.7 opqrs	4.67 hijkl	21.7	19.2 bcdef	55.5 klmn	96.4 fgh
11.5 cdefgh	4.19 cdefghij	3.9	3.9 abc	25.5 abcdefghijk	33.2 abcd
14.4 mnopqrs	4.01 bcdefghi	17.4	24.8 efgh	46.3 ghijklmn	88.4 efgh
14.3 mnopqr	5.01 l	40.3	43.7 jk	49.0 ijklmn	132.9 hijk
14.6 nopqrs	4.24 cdefghijk	24.2	22.3 defg	47.7 hijklmn	94.2 fgh
16.1 st	4.89 jkl	25.2	26.4 fghi	43.3 efghijklm	95.0 fgh
14.7 opqrs	4.32 cdefghijkl	48.4	20.5 cdefg	53.5 klmn	122.5 ghij
15.7 qrst	4.61 fghijkl	66.3	29.8 fghij	64.3 mno	160.3 jkl
15.9 rst	4.63 ghijkl	49.3	26.0 fghi	61.1 mno	136.4 hijk
12.4 ghijkl	3.90 abcdef	8.8	7.5 abcde	45.6 fghijklmn	61.9 bcdef
13.8 klmnop	4.08 bcdefghi	22.2	18.1 abcdef	27.6 abcdefghijkl	67.7 cdef
12.8 hijklm	4.43 defghijk	30.7	31.9 fghijk	25.5 abcdefghijk	88.1 efgh
14.5 mnopqrs	4.71 ijkl	25.4	32.6 fghijk	52.8 jklmn	110.8 fghij
16.6 t	4.33 cdefghijkl	31.8	43.1 ijk	55.8 klmn	130.5 hij
13.5 jklmnop	4.14 bcdefghi	55.3	41.6 hijk	93.2 o	190.1 l
16.7 t	4.50 efghijkl	47.9	28.7 fghij	32.2 abcdefghijklm	108.8 ghij
15.2 pqrst	4.95 kl	59.9	47.3 k	73.6 no	180.3 kl
14.1 lmnopq	4.22 cdefghijk	4.9	4.6 abc	13.3 abcdef	23.0 abc
11.0 bcdefg	3.96 bcdefgh	8.8	3.9 abc	25.4 abcdefghijk	38.1 abcd
9.4 ab	3.67 abc	2.6	0.0 a	1.3 a	3.9 a
11.9 defghij	4.32 cdefghijkl	15.3	1.4 a	25.4 abcdefghijk	42.1 abcde
11.0 bcdefg	3.75 abcd	1.2	6.6 abcd	5.6 ab	13.7 ab
9.9 abc	3.55 abc	0.0	0.0 a	5.6 ab	5.6 a
11.6 cdefghi	3.65 abc	1.2	3.6 abc	1.2 a	6.0 a
9.2 a	4.03 bcdefghi	1.5	0.0 a	7.4 abc	8.8 a
11.3 cdefgh	3.42 ab	0.0	0.01 a	1.5 a	1.5 a
10.6 abcdef	3.60 abc	6.1	0.0 a	11.3 abcde	17.4 ab
11.0 bcdefg	3.66 abc	8.8	0.0 a	5.9 ab	14.7 ab
12.3 fghijkl	3.75 abcd	3.7	0.0 a	25.8 abcdefghijk	29.5 abcd
10.8 abcdefg	4.00 bcdefghi	11.3	0.0 a	14.1 abcdefg	25.4 abcd
9.4 ab	3.86 abcde	0.0	0.0 a	3.7 ab	3.7 a
11.3 cdefgh	4.42 efghijkl	5.3	1.3 a	7.1 abc	13.7 ab
10.8 abcdefg	3.23 a	0.0	0.0 a	1.7 a	1.7 a
12.0 efghij	4.04 bcdefghi	3.7	1.2 a	15.3 abcdefgh	20.2 abc
12.0 efghij	3.78 abcde	6.5	1.7 a	10.4 abcd	18.6 abc
9.3 ab	3.41 ab	0.0	1.5 a	11.4 abcde	13.9 a
12.9 hijklmn	4.14 bcdefghi	1.4	1.4 a	27.2 abcdefghijkl	30.0 abcd
10.2 abcd	3.86 abcde	3.3	3.0 ab	20.4 abcdefghij	26.8 abcd
10.5 abcde	3.92 abcdefg	0.0	0.0 a	0.0 a	0.0 a
12.0 efghij	3.93 abcdefg	1.3	2.7 ab	18.2 abcdefghi	22.2 abc
19.6 abcdef	3.89 abcdef	0.0	0.0 a	1.6 a	1.6 a
12.3 fghijkl	4.25 cdefghijk	5.8	1.5 a	7.9 abc	15.2 ab
*	*	n.s.	*	n.s.	*
**	**	n.s.	*	*	*



according to VICTOR's (1948) observation, that the time of anthesis was correlated particularly with growth in treatments showing the highest degree of unbalance, where a marked delay in anthesis occurred on plants definitely stunted (in our case the treatments 1 : 1 : 1 and 2 : 2 : 2, as observed previously).

This would indicate that besides the total P uptake and % content the time and quantity of flowering were related to the physiological age of the plants.

The data obtained with regard to loamy sand soil indicated that all NPK combinations were not statistically effective either on P behaviour in plants or on flowering. In spite of the nutrient combination 1 : 2 : 1 being superior to other treatments, this favourable effect was possibly due to the effect of phosphate in decreasing soil strength (KIRK 1945, LUTZ *et al.* 1966) which was associated with better root growth and higher P uptake. This action was attributed to the replacement of OH ions by phosphate ions on the edge of the clay particles causing aggregation. If treatment 1 : 2 : 1 (26.1% and 33.8% of opened and total flowers, respectively) was compared with treatment 2 : 2 : 1 (3.9% and 4.0% of opened and total flowers, respectively) the depressive effect of an increasing N to P ratio in loamy sand could be shown; the opposite was observed in sandy soil.

*Effect of seed treatments  $\times$  nutrient ratio.* The results showed that, in general, in spite of the fact that fresh and dry weights of seedlings, dry weight of root, stem length II and opened flower did not seem to be affected by seed treatments when combined with the available nutrient ratios or quantities, there appeared to be a relatively similar trend in both stem length and diameter as well as in set and total flowers, which were significantly affected (Table 3). With the three seed treatments the nutrient ratio 2 : 1 : 2 seemed to be the most effective on stem length and diameter when compared with their respective ratios. No significant differences were obtained between the mentioned ratio and the ratios 1 : 2 : 1 and 1 : 2 : 2 when combined with pre-sowing seed treatments regarding the stem length.

In respect of flowering, with both dry seeds and seeds soaked in water, and for the combinations 2 : 1 : 1 and 2 : 2 : 1 for seeds soaked in Volldünger no significant differences were found, though treated seeds were significantly superior to the dry ones. Seed soaking as well as nutrients had a stimulating effect, as reported already in this study and by other authors (ANISIMOV—NUSKOVA 1973, NIKOLOV 1973). It was shown that pre-sowing treatments improved the nitrogen utilization of beans grown on poor soil fertilized with ammonium nitrate. On the other hand, the combinations 1 : 1 : 1 and 2 : 2 : 2 seemed to be the most depressive treatments for flowering when interacting with the three seed treatments, though with seeds soaked in water the injury was lower.

*Effect of seed treatments  $\times$  soil type.* Table 2 shows that the effect of seed treatments combined with soil type on stem length II appears to be more consistent than on other seedling performance factors or on flowering, the interaction of soaked seeds and sandy soil being the most effective. This might be explained by the favourable effect of both sandy soil and soaking of seeds, as already mentioned.

*Effect of soil type  $\times$  seed treatments  $\times$  nutrients.* Irrespective of dry weight of seedlings and flowering abscission, which were not significantly affected by the interaction of soil type, seed treatments and nutrients, the fresh weight of seedlings, stem length, diameter and flowering were significantly affected (Table 3). It can be concluded, in general, that sandy soil that contains the ratio 2 : 1 : 2 was generally more effective in stimulating the growth of seedlings with the three seed treatments. But when the ratio of N : P<sub>2</sub>O<sub>5</sub> : K<sub>2</sub>O was 2 : 2 : 1 or 1 : 2 : 2, then the presoaked seeds were more favourable than the dry ones.

With respect to loamy sand, no clear effect was obtained and the effect of treatments on growth characters fluctuated from one to another. In spite of this, it can be concluded that the nutrient ratio 1 : 2 : 1 with the three seed treatments, the ratios 1 : 1 : 1 or 2 : 2 : 2 with seed soaked in Volldünger and the ratios 2 : 1 : 2 or 2 : 2 : 2 with seed soaked in water

seemed to be the best treatments for the production of good seedlings with the best growth quality (stem length and diameter).

Regarding flowering, correlation coefficients and linear regressions were worked out as effects by the interaction of soil types  $\times$  seed treatments  $\times$  available nutrient ratios in the soil for the relationship between some seedling characters summarized as follows:

Summary of "r" values for linear regression equations for fresh weight/seedling, P uptake and P % in seedling top as independent variables (X) and flower set %, opened flower % and total flower % as dependent variables (Y).

X \ Y	Y		
	Fl. set %	Opened fl. %	Total fl. %
Fresh wt.	0.80***	0.77***	0.83***
P uptake	0.76***	0.80***	0.80***
P %	0.40**	0.43**	0.42**

"r" value needed for significance at 1%

\*\* = 0.35 and at 0.1%; \*\*\* = 0.44, P = 48.

The data reveal that all the dependent characters studied increased as the respective independent character increased. This was an attempt to interpret some of the phenomena resulting from the interaction of the factors studied.

Moreover, from Table 3 it can be seen that in sandy soil, the nutrient combination ratios 2 : 1 : 1 and 2 : 1 : 2 with dry seeds, the ratios 1 : 1 : 2, 2 : 1 : 2 and 2 : 2 : 1 with seeds soaked in water and the ratios 2 : 1 : 1 and 2 : 2 : 1 with seeds soaked in Volldünger were generally the most effective treatments in spite of their not differing significantly within themselves. The treatment 2 : 1 : 1 with seeds soaked in Volldünger produced relatively higher numbers of flowers. On the other hand, in loamy sand soil there were no statistically significant differences either among or within the treatments. In spite of this, 1 : 2 : 1 and 1 : 2 : 2 seemed to be the most favourable.

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### References

- ANGELOV, L. (1974): The effect of mineral fertilizers on certain features of determinate and indeterminate tomato cultivars. *Gradinarska i Lozarska*, 11/1, 71-76.
- ANISIMOV, A. A.—NUSKOVA, E. L. (1973): Effect of Zn and Co on ATP-ase activity of fodder beet. *Uchenye Zapiski. Gor'kovskii Universitet, Biologicheskaya*, 178, 52-57.
- BELOKOBYL'SKÜ, L. M.—MALYKIRA, V. F. (1970): Effect of manganese sulphate on lysine percentage in the protein of maize grain. *Trudy Vorshlovgradskogo Sel'skokhozyaistvennogo Instituta*, 20, 11-12.
- BERTRAND, A. R.—KOHNE, H. (1957): Subsoil conditions and their effects on oxygen supply and the growth of corn roots. *Soil Sci. Soc. Am. Proc.*, 21, 135-140.
- BESKROVNAYA, V. N. (1970): The effect of presowing treatment of seeds with micro-elements on the intensity of fruit formation in tomatoes. *Trudy Kamenets-Podol'skogo Sel'skokhozyaistvennogo Instituta*, 15, 100-104.



- EL-SAWAH, M. H. (1982): Influence of soil types, pre-sowing seed treatments, available amounts of soil nutrients and their combinations on tomatoes. II. Effect on mineral status of seedlings and leaf pigment contents. *Acta Agron. Hung.*, **32**, 392-399.
- FERENCZ, V.—SOMOS, A.—MÁRKUS, L. (1964): Paradicsom növény tápanyagforgalmának néhány összefüggése. A táplálékanyagok különböző arányának hatása a növény fejlődésére és terméshozamára (Some correlations of nutrient balance in tomato plants. Effect of different proportions of nutrients on plant development and yield). *Agrokémia és Talajtan*, **13**, 205-218.
- GAAROV, B. KH. (1971): The effect of seed hardening on some physiological indices and yield of tomato. *Nauchn. Trudü Samarkandskir Sel'skokhozyaistvennogo Instituta*, **12**, 25-40.
- KIRK, L. (1945): The influence of soil aeration on the growth and absorption of nutrients by corn plants. *Soil Sci. Soc. Proc.*, **9**, 263-268.
- LUTZ, J. F.—PINTO, R. A.—RICARDO, G.—HITTON, H. G. (1966): Effect of phosphorus on some physical properties of soil: II. Water retention. *Soil Sci. Soc. Amer. Proc.*, **30**, 433-437.
- NIKOLOV, B. A. (1973): The role of Mo in nitrogen assimilation by beans fertilized with ammonium nitrate. *Pochvoznanie i Agrokimiya*, **8**, 95-103.
- PHILLIPS, R. E.—KIRKHAM, D. (1962): Mechanical impedance and corn seedling root growth. *Soil Sci. Soc. Amer. Proc.*, **26**, 319-322.
- PROHÁSZKA, K.—HAMAR, N. (1977): Effect of nutrition on the nutrient composition in different parts of tomatoes. *Acta Agron. Hung.*, **26**, 167-176.
- STEINER, A. A. (1966): The influence of the chemical composition of a nutrient solution on the production of tomato plants. *Plant and Soil*, **24**, 454-466.
- TAYLOR, H. M.—HERBERT, R. G. (1963): Penetration of cotton seedling taproots as influenced by bulk density, moisture content, and strength of soil. *Soil Sci.*, **96**, 153-156.
- TROUSE, A. C. JR.—BAVER, L. D. (1962): The effect of soil compaction on root development. *Trans. Joint Meeting Com. IV and V. Intern. Soc. Soil Sci.*, New Zealand, 253-263.
- UZO, J. O. (1971): Effect of N, P and K on the yield of tomato in the humid tropics. *Hort. Res.*, **11/2**, 65-74.
- VEIHMEYER, I.—HENDRICKSON, A. H. (1948): Soil density and root penetration. *Soil Sci.*, **65**, 487-493.
- VICTOR, N. L. (1948): Nutrient-element balance and time of anthesis in tomato flowers. *Proc. Amer. Soc. Hort. Sci.*, **52**, 347-349.
- VLASYUK, P. A.—MATYUSHENKO, A. V.—BLAGANS'KA, V. E.—GERYACHOVA, L. O. (1974): Effect of Mo, Zn and Mn on yield and fractional composition of proteins of grains of maize. *Visr. Sil-hospod. Nauky*, **10**, 38-41.
- WOODRUFF, D. R. (1969): Studies on presowing drought hardening of wheat. *Aust. J. Agric. Res.*, **20**, 13-24.

#### EFFECT OF GYPSUM APPLICATION ON AMELIORATION OF SODIC SOILS CROPPED WITH RICE

An estimated seven million hectares of land in India suffers either from an excess of soluble salts, or excess sodium in the exchange complex, or both (ABROL—BHUMBLA 1971). The amelioration of sodic soils requires the replacement of the exchangeable Na with cations such as Ca, which lower the pH and improve the physical condition of the soil. The soluble salts and Na must also be removed from the soil profile by drainage water. The most readily available and widely used source of calcium for replacing Na ions is gypsum, and rice is the most suitable crop to be grown (KANWAR—BHUMBLA 1969, ABROL *et al.* 1973). However, the amount of amendment needed for amelioration depends on the soil characteristics, the crop to be grown, the rate of replacement, etc.

The objective of this investigation was to study the effect of different rates of gypsum on soil improvement and on the yields of paddy and subsequent other crops grown on sodic soils.

In a field experiment at the Agricultural Research Station of the University of Udaipur, Vallabh Nagar, the efficiency of different levels of gypsum application for soil improvement



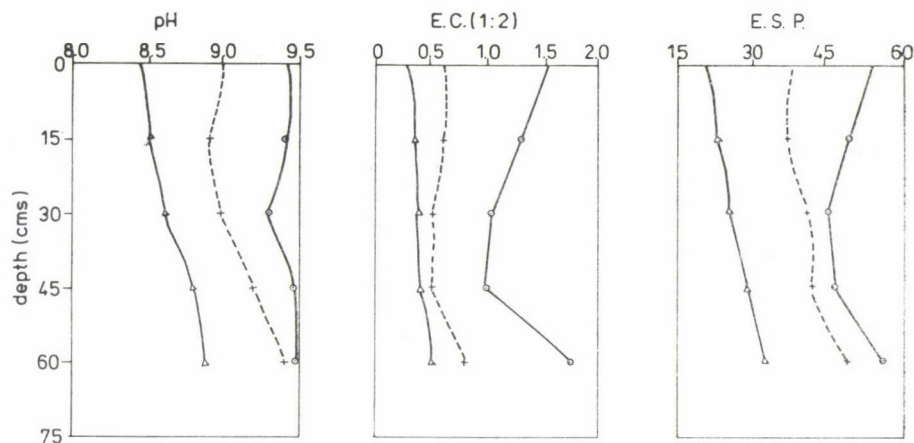


Fig. 1. Effect of gypsum treatments on pH, E.C. and E.S.P. of soil (initial value ·····; after paddy with no gypsum ----×----×---- after paddy with 2.5 tons of gypsum ---)

and crop production on a sodic soil was examined. The soil of the experimental plots was clay loam in texture, having a pH of 9.4 and an ESP value of 49.8. Five gypsum treatments, 0.0, 2.5, 5.0, 7.5 and 10.0 tons/ha (equivalent to 0, 25, 50, 75 and 100% of gypsum requirement for the 15 cm of soil) were replicated three times in a randomized block design. The plot size was 15 × 8 m. After incorporating gypsum into the upper 15 cm of soil, 35-day-old seedlings of Zaya paddy were transplanted in the Kharif season 1975 with 2–3 seedlings per hill. During the Rabi season 1975–76, wheat, barley and oats were grown on the same plots, subdivided into 5 × 8 m sub-plots. The crop sequence for 1976–77 was sorghum in Kharif and oats in Rabi, both as fodder crops. All the crops received a basal dose of N, P and K and the recommended cultural practices were followed. Soil samples were collected from the plots at various depths before the addition of the amendment and after harvesting the paddy crop, and were analysed for pH, EC and ESP.

The results of the present investigation on the effect of gypsum on soil properties and crop yield are presented below:

**Effect on soil pH, soluble salts and ESP.** Analysis of soil samples taken from various depths after harvesting the paddy crop showed a marked decrease in the pH and ESP of the soils due to the treatments, as compared to the initial values (Table 1, Fig. 1). The pH of the untreated control plot decreased from 9.4 to 8.95, and was further reduced to 8.54 with gypsum application. The exchangeable sodium percentage for the upper 15 cm soil was also noticeably reduced from 49.4 to 22.52 as the result of 2.5 tons of gypsum application. A decrease in pH and ESP in the lower soil depth was also noticed. The lowering of pH and ESP was probably due to the fact that the Ca in the gypsum exchanged Na from the soil colloids, while the rice crop growth also helped in soil amelioration by increasing the volume of water drained and the solubilization of  $\text{CaCO}_3$  through root effects (CHHABRA—ABROL 1977, SHAHI *et al.* 1978). The soluble salts in the soil profile were reduced markedly even by growing rice and the values were reduced further by the application of gypsum. More than 50% of the salt initially present was displaced from the soil profile by rice growing and the addition of gypsum. No appreciable change in the pH and EC of the soil was noted when applying more than 2.5 tons of gypsum; however, the values tended to decrease slightly with the rate of gypsum applied.

**Table 1**  
*Effect of gypsum treatments on pH, E.C. and ESP of soil*

Depth (cm)	pH			E.C. (1 : 2)			ESP		
	Initial value	After paddy with no gypsum	After paddy with 2.5 tons of gypsum	Initial value	After paddy with no gypsum	After paddy with 2.5 tons of gypsum	Initial value	After paddy with no gypsum	After paddy with 2.5 tons of gypsum
0-15	9.4	8.9	8.52	1.27	0.63	0.36	49.4	37.6	22.5
15-30	9.3	9.0	8.63	1.00	0.50	0.40	46.0	40.0	24.1
30-45	9.5	9.2	8.80	0.95	0.49	0.46	47.3	41.0	29.1
45-60	9.5	9.4	8.95	1.75	0.79	0.53	55.6	48.0	32.0

**Table 2**  
*Effect of different levels of gypsum application on crop yields*

Levels of gypsum (tons/ha)	Grain yields (q/ha) during 1975-76				Fodder yields (q/ha) during 1976-77	
	Paddy	Wheat	Barley	Oats	Jowar	Oats
0.	40.37	20.75	22.58	20.00	225.8	355.8
2.5	54.65	29.92	31.07	24.98	231.7	383.7
5.0	58.03	33.25	34.17	27.67	267.9	404.6
7.5	59.03	34.42	38.08	28.50	261.2	394.6
10.0	60.55	36.00	36.25	28.00	262.9	398.3
C.D. at 5% level	4.09	6.98	7.10	4.13	[n.s.	n.s.
C.V.	7.62	12.27	12.03	19.93	12.01	8.66

*Effect on crop yield.* The grain yields of different crops, as influenced by rates of gypsum application, are presented in Table 2. The table shows that the yield of paddy, wheat, barley and oats increased significantly with an addition of gypsum, but the percentage increase varied with the crop. A 35.3% increase in grain yield was obtained for paddy with the application of 2.5 tons/ha of gypsum and this was significantly higher than the control. At higher levels, though the yield increased, the yield differences above 2.5 tons/ha of gypsum were not significant. Yields of Rabi crops were also increased considerably during 1975-76 as the result of applying gypsum to the previous paddy crop. At 2.5 tons/ha of gypsum, the grain yields of wheat, barley and oats increased by 44.9, 37.5 and 24.9% respectively as compared to the control. No significant increase was observed in the yields of these crops at over 2.5 tons of gypsum; however, the yield tended to increase with the amount of gypsum applied. A very marginal increase in the yield of crops was noted beyond an application of 5 tons of gypsum. The fodder yield of both Jowar and oats obtained during the year 1976-77 on treated plots was also higher than the yield obtained on control plots, but the differences due to various treatments were non-significant. The results of the present investigation therefore indicate that the application of 2.5 to 5.0 tons/ha of gypsum (equivalent to 25 to 50% of the gypsum requirement) was adequate to obtain optimum grain yields of paddy, wheat, barley and oats.

Increased yields may be due to an improvement in soil conditions brought about by the application of gypsum and the growing of rice, which lowered the pH, salt content and ESP of the soil. The results are in agreement with those obtained by KANWAR—CHAWLA (1963), ABROL—BHUMBLA (1971) and BHUMBLA (1972), who observed an improvement in soil properties and optimum crop yield on sodic soils due to gypsum application at 30 to 50% levels of gypsum requirement.

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### References

- ABROL, I. P.—BHUMBLA, D. R. (1971): Saline and alkali soils in India — their occurrence and management. World Soil Resources. F.A.O. Report No. 41, 42–51.
- ABROL, I. P.—DARGAN, K. S.—BHUMBLA, D. R. (1973): Reclaiming alkali soils. Bull. No. 2, CSSRI, Karnal, India.
- BHUMBLA, D. R. (1972): Reclamation and utilisation of salt affected soils. Indian Farming, 22/2, 19–21.
- CHHABRA, R.—ABROL, I. P. (1977): Reclaiming effect of rice grown in sodic soils. Soil Sci., 124, 49–55.
- KANWAR, J. S.—CHAWLA, V. K. (1963): Comparative study of the effect of gypsum and pressmud on physico-chemical properties of saline-alkali soils. J. Soil Water Cons. India, 11, 95–106.
- KANWAR, J. S.—BHUMBLA, D. R. (1969): Physico-chemical characteristics of sodic soils of the Punjab and Haryana and their amelioration by use of gypsum. Agrochem. Talaj., 18, 315–320.
- SHAH, H. N.—MOSKINA, M. S.—GILL, P. S. (1978): Effect of different levels of gypsum application on soil characteristics and growth and yield of rice. Plant and Soil, 49, 437–442.

### PHOSPHORUS ADSORPTION IN RELATION TO IMPORTANT SOIL PROPERTIES IN SOME ACID SOILS OF NORTH-WEST INDIA

The acid soils of Himachal Pradesh province in North-West India have been reported to exhibit phosphorus deficiency to a fairly large extent (RAMAMOORTHY—BAJAJ 1969, SHARMA—BHUMBLA 1975) and to respond to a heavy application of phosphorus (MAHAJAN *et al.* 1975), which suggests the extensive retention of added phosphorus in these soils. Although several factors have been found to be responsible for the adsorption of added phosphorus in acid soils in various parts of the world (KANWAR 1956, RENNIE—MCKERCHER 1959, SAUNDERS 1965, AHENKORAH 1968, KUO—LOTSE 1974, VIJAY CHANDRAN—HARTAR 1975), there is a lack of scientific information on this aspect of acid soils in India as a whole, since the earlier work on phosphate adsorption in this country was confined to saline and alkali soils (SUBRAMANIAN 1965, KANWAR—RAYCHAUDHARY 1971, VIJ-DEV 1978). The present investigation was, therefore, undertaken to study the extent and mechanism of phosphate adsorption in a few representative acid soils of Himachal Pradesh province in North-West India, in relation to their important properties.

Surface soil samples from a depth of 0–15 cm were collected from five representative places, i.e. Palampur, Nagrota, Andretta, Kangra and Hamirpur, which are located in an agriculturally very important belt exhibiting the problem of soil acidity in the Himachal Pradesh province of North-West India. The first four soils, namely Palampur, Nagrota,



Andretta and Kangra, have developed under a temperate, humid climate in a coniferous vegetation and their parent material is composed of slates, phyllites, quartzite, schist, gneiss and granites. They fall under the suborder "alfisols" and their annual rainfall exceeds 1500 mm. The fifth soil, however, i.e. Hamirpur, owes its origin to sand and silt stone and to calcareous conglomerates (shales). The annual rainfall in this soil ranges between 750–1500 mm. Paddy, maize and wheat are the most important cereal crops being grown on both groups of soils. The soil samples were air dried, ground and passed through a 2 mm sieve. The pH of the samples was determined using a 1 : 2 soil/water suspension with a Beckman pH meter. Clay and sesquioxides were determined as outlined by PIPER (1950). The adsorption of applied phosphate was studied by taking a 1 g sample of each soil and equilibrating it for 24 hr at room temperature with 25 ml of  $\text{KH}_2\text{PO}_4$  solution (pH 7) in the concentration range varying from 0 to 150 ppm P, i.e. 0 to 150  $\mu\text{g}$  P/g soil. The final concentration of phosphorus in the supernatant was determined by the method of OLSEN *et al.* (1954). The amount of phosphorus adsorbed was calculated as the difference between the initial and final concentrations of phosphorus in each solution. The simple co-efficients of correlation between the adsorption parameter ( $k$ ) of the Freundlich adsorption equation and the soil properties, were computed by the method proposed by SNEDECOR—COCHRAN (1968).

The effect of time on the extent of adsorption was studied in a number of samples, and a period of 24 hr was found to result in maximum adsorption of phosphorus. Hence, the time of shaking was taken as 24 hr in this experiment. The physico-chemical properties of the soils, and Freundlich constants  $k$  and  $n$ , are listed in Table 1. The adsorption data are presented in Table 2, while the simple coefficients of correlation between the adsorption parameter ( $k$ ) of the Freundlich adsorption equation and the soil properties, are given in Table 3. The Freundlich adsorption isotherms of phosphorus in various soils are diagrammatically illustrated in Fig. 1. It was observed that the adsorption data presented in Table 2 fitted the logarithmic form of the Freundlich adsorption equation,  $\log x/m = \log k + \frac{1}{n} \log C$ ; where  $x/m$  is the amount of phosphorus adsorbed per unit weight of soil ( $\mu\text{g/g}$ ) and  $C$  is the equilibrium phosphorus concentration in the solution phase ( $\mu\text{g/ml}$ ). The Freundlich constant  $k$  characterizes the relative P adsorption capacity, whereas  $n$ , which is also a constant, signifies the degree of non-linearity between the solution concentration and adsorption. The values of  $k$  and  $n$  were computed from the intercepts and the slopes of the respective adsorption isotherms (Fig. 1), plotted between the logarithm of the amount of P adsorbed ( $\mu\text{g/g}$ ) and the logarithm of the equilibrium solution concentration of P ( $\mu\text{g/ml}$ ). A look at the phosphate adsorption isotherms (Fig. 1) indicates a linear relationship between  $\log x/m$  and  $\log C$ .

Since the magnitude of the Freundlich constant ( $k$ ) is indicative of the extent of adsorption, the data presented in Table 1 show that the adsorption capacity of soils for applied phosphorus follows the order Palampur > Nagrota > Andretta > Kangra > Hamirpur. The soils of Palampur, Nagrota, Andretta and Kangra recorded a considerably higher adsorption of phosphorus compared to that of Hamirpur soil. The results further indicate that the adsorption capacity of the soils is related to pH, sesquioxides and organic matter (Table 3). Thus, the soils of Palampur, Nagrota, Andretta and Kangra, which have low pH and are rich in sesquioxides and organic matter, have higher phosphorus adsorption capacity than the soil of Hamirpur. For instance, at an initial concentration of 50 ppm of added P, the soils of Palampur, Nagrota, Andretta and Kangra adsorbed 82, 80.4, 77.4 and 75% respectively of added P, whereas the soil of Hamirpur adsorbed only 55% (Table 2).

An attempt was also made to assess the relationship between relative P adsorption capacity ( $k$ ) and the soil properties through simple co-efficients of correlation, which are given in Table 3. A perusal of this table indicates that the Freundlich constant ( $k$ ), which is a parameter of P adsorption capacity, had a significant positive association with sesquioxides ( $r = +0.81, p = 0.05$ ) whereas it had a significant negative association with pH ( $r = -0.72$ ,

**Table 1***Physico-chemical properties and Freundlich constants of soils*

Soils	pH 1:2 Soil : water	Clay	Organic matter %	Sesquioxides	Freundlich constants	
					k	n
Palampur	5.5	26.5	2.0	0.95	1.90	0.68
Nagrota	5.3	17.6	2.1	1.12	1.58	0.75
Andretta	4.9	25.4	2.0	1.15	1.20	0.65
Kangra	5.9	20.4	1.4	0.85	1.04	0.66
Hamirpur	6.5	17.6	1.0	0.29	0.30	0.87

**Table 2***Adsorption data of added phosphorus in five soils*

P added ( $\mu\text{g/ml}$ )	Amount of P absorbed ( $\mu\text{g/g}$ ) ( $x/m$ )	Equilibrium concentration of P in solution ( $\mu\text{g/ml}$ ) ( $C$ )	Log ( $C$ )	Log ( $x/m$ )
<i>Palampur pH 5.5</i>				
50	41.0	9.0	0.95	1.61
75	62.9	12.1	1.08	1.80
100	84.5	15.5	1.19	1.93
150	129.7	20.3	1.30	2.11
<i>Nagrota pH 5.3</i>				
50	40.2	9.8	0.99	1.60
75	61.6	13.4	1.12	1.79
100	82.5	17.5	1.24	1.91
150	126.5	23.5	1.37	2.10
<i>Andretta pH 4.9</i>				
50	38.7	11.3	1.05	1.58
75	61.6	13.4	1.12	1.79
100	83.0	17.0	1.23	1.92
150	129.3	20.7	1.31	2.11
<i>Kangra pH 5.9</i>				
50	37.5	12.5	1.09	1.57
75	60.0	15.0	1.17	1.78
100	79.3	20.7	1.31	1.90
150	126.6	23.6	1.37	2.10
<i>Hamirpur pH 6.5</i>				
50	27.5	22.5	1.35	1.44
75	44.6	30.4	1.48	1.65
100	62.5	37.5	1.57	1.79
150	97.2	52.8	1.72	1.98

$p = 0.1$ ). However, the adsorption capacity ( $k$ ) had a significant positive relationship with organic matter ( $r = +0.89$ ,  $P = 0.05$ ).

The fact that the soils of Palampur, Nagrota, Andretta and Kangra adsorbed 82, 80.4, 77.4 and 75% respectively of added P at an initial concentration of 50 ppm P, while Hamirpur soil adsorbed 55%, is in conformity with the findings of RENNIE—MCKERCHER (1959) for some acid soils of Canada, SAUNDERS (1965) for acid soils of New Zealand and KUO—LOTSE (1974) for some acidic lake sediments of North America. In the soils under investigation, the free sesquioxides seem to be a major contributory factor towards the adsorption of phosphate. This is evident from the significant positive correlation ( $r = 0.81$ ,  $p = 0.05$ ) between the Freundlich constant ( $k$ ) and the sesquioxides. KANWAR (1956), SAUNDERS

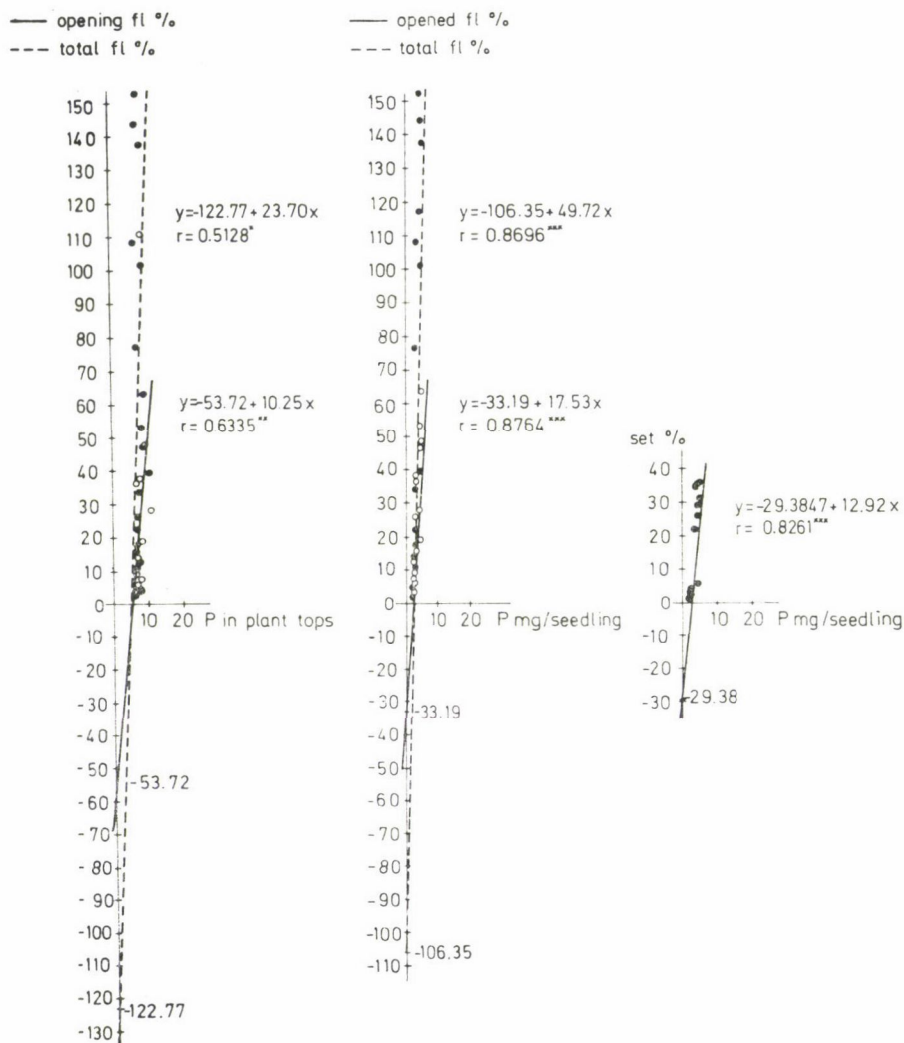


Fig. 1. Freundlich adsorption isotherms of phosphorus in various soils



**Table 3**  
Coefficients of correlation between  
Freundlich constant ( $k$ )  
and soil properties

Soil property	' $r$ '
1. pH	-0.72*
2. Organic matter	+0.89**
3. Clay	+0.56
4. Sesquioxides	+0.81**
5. Organic matter vs. sesquioxides	+0.93***

\* Significant at 10% level.

\*\* Significant at 5% level.

\*\*\* Significant at 1% level.

(1965) and GEBHARDT—COLEMAN (1974) also observed high-affinity adsorption for phosphate on the surface of sesquioxides. The dominance of sesquioxides in most of the soils used here is probably due to low pH and intense weathering, resulting from high rainfall (1500 mm). The negative correlation between pH and the phosphate adsorption capacity  $k$  ( $r = 0.72$ ,  $p = 0.1$ ) indicates a decline in P adsorption with an increase in pH from the acidic to the neutral range. This would be anticipated from the greater activity of sesquioxides at lower pH, which eventually contributes indirectly to P adsorption. The pH has also been shown to be negatively correlated with P adsorption by UDO—UZU (1972) in their studies on acidic Nigerian soils. The significant positive relationship found between phosphorus adsorption capacity ( $k$ ) and organic matter ( $r = +0.89$ ,  $p = 0.05$ ) in the present study, is in accordance with the findings of BROOMFIELD (1965), SAINI—MCLEAN (1965), HARTAR (1969), SYERS *et al.* (1971) and VIJAY CHANDRAN—HARTAR (1975). SAINI—MCLEAN (1965) explained it on the basis of the association of organic matter with aluminium and iron (sesquioxides) in the form of complexes which are very reactive in phosphorus retention, while VIJAY CHANDRAN—HARTAR (1975) attributed this behaviour to the presence of anion adsorption sites on the organic matter itself. In the present study, the organic matter was found to be related to sesquioxides ( $r = +0.93$ ,  $p = 0.01$ ), indicating that the major portion of the sesquioxides has been chelated by organic matter. Therefore, the role played by the organic matter in the adsorption of phosphorus in the present study is probably due to its association with sesquioxides. Although clay was found to be positively associated with phosphorus adsorption capacity, the correlation was not significant. Thus, clay as such, has not contributed much to the adsorption of phosphorus in these soils. SAINI—MCLEAN (1965) and AHENKORAH (1968) also observed similar results for New Brunswick soils in Canada and cocoa-growing soils of Ghana, respectively.

The mechanism of phosphorus adsorption in the present soils thus appears to operate through the presence of sesquioxides and organic matter. On hydrolysis, the sesquioxides expose a much larger specific surface due to their colloidal nature. High-affinity adsorption of phosphorus takes place on protonated sites, along with low-affinity adsorption of potassium from  $\text{KH}_2\text{PO}_4$  and the formation of variscite-like substances, as suggested by HINGSTON *et al.* (1967) and GEBHARDT—COLEMAN (1974).

There is also the possibility of hydroxyl (OH-) displacement by  $\text{H}_2\text{PO}_4^-$  and a subsequent reaction between potassium and weakly exchanging materials as proposed by MULJADI *et al.* (1966) and GEBHARDT—COLEMAN (1974) in such soils as are rich in sesquioxides. Since the soils under study indicate a high degree of correlation between sesquioxides and phosphorus adsorption capacity, both these mechanisms seem to be possible. The organic matter is perhaps involved in phosphorus adsorption due to its association with sesquioxides in these soils.

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### References

- AHENKORAH, Y. (1968): Phosphorus retention capacities of some cocoa growing soils of Ghana and their relationship with soil properties. *Soil Sci.*, **105**, 24–30.
- BROMFIELD, S. M. (1965): Studies on the relative importance of iron and aluminium in the sorption of phosphate by some Australian soils. *Australian J. soil Res.*, **3**, 31–44.
- GEBHARDT, H.—COLEMAN, N. T. (1974): Anion adsorption by allophanic tropical soils. III. Phosphate adsorption. *Soil Sci. Soc. Amer. Proc.*, **38**, 263–266.
- HARTAR, R. D. (1969): Phosphorus adsorption sites in soils. *Soil Sci. Soc. Amer. Proc.*, **33**, 630–631.
- HINGSTON, F. J.—ATKINSON, R. J.—POSNAR, A. M.—QUIRK, J. P. (1967): Specific adsorption of anions. *Nature*, **215**, 1459–1461.
- KANWAR, J. S. (1956): Phosphorus retention in some Australian soils. *Soil Sci.*, **82**, 43–50.
- KANWAR, J. S.—RAYCHAUDHARY, R. S. (1971): Review of soil research in India. *Indian Soc. Soil Sci. IARI*, New Delhi (India).
- KUO, S.—LOTSE, E. G. (1974): Kinetics of phosphate adsorption and desorption by lake sediments. *Soil Sci. Soc. Amer. Proc.*, **38**, 50–54.
- MAHAJAN, K. K.—TRIPATHI, B. R.—KANWAR, B. S. (1975): Response of wheat crop to different sources of phosphorus on grey brown podzolic soils. *Himachal J. Agri Res.*, **2**, 33–37.
- MULJADI, D. A.—POSNAR, M.—QUIRK, J. P. (1966): The mechanism of phosphate adsorption by kaolinite, gibbsite and pseudoboehmite (I–III). *J. Soil Sci.*, **17**, 212–247.
- OLSEN, S. R.—COLE, C. V.—WATANABE, F. S.—DEAN, L. A. (1954): Estimation of available phosphorus in soils with  $\text{NaHCO}_3$ . *USDA Circ.* 939.
- PIPER, C. S. (1950): *Soil and Plant Analysis*. Inter Science Publishers Inc., New York (USA).
- RAMAMOORTHY, B.—BAJAJ, J. C. (1969): Available nitrogen, phosphorus and potassium status of Indian soils. *Fertilizer News*, **14**, 1–12.
- RENNIE, D. A.—MCKERCHER, R. B. (1959): Adsorption of phosphorus by four Saskatchewan soils. *Soil Sci.*, **91**, 357–359.
- SAINI, G. R.—MCLEAN, A. J. (1965): Phosphorus retention capacities of some New Brunswick soils and their relationship with soil properties. *Can. J. Soil Sci.*, **45**, 15–18.
- SAUNDERS, W. M. H. (1965): Phosphate retention by New Zealand soils and its relationship to free sesquioxides, organic matter and other soil properties. *N.Z.J. Agr. Res.*, **8**, 30–37.
- SHARMA, P. K.—BHUMBLA, D. R. (1975): Available phosphorus distribution in the soils of Kangra and Kulu districts of Himachal Pradesh and comparison of extraction procedures. *Himachal J. Agri. Res.*, **3**, 1–8.
- SNEDECOR, G. W.—COCHRAN, W. G. (1968): *Statistical Methods*. Oxford and IBH Publishing Co., New Delhi (India).
- SUBRAMANIAN, T. R. (1965): Studies on exchangeable P in soils and clay minerals. *Indian J. Agr. Sci.*, **35**, 79–84.
- SYERS, J. K.—EVANS, T. D.—WILLIAMS, J. D. H.—MURDOCK, J. T. (1971): Phosphate sorption parameters of representative soils from Rio Grande do Sul, Brazil. *Soil Sci.*, **112**, 267–275.



- UDO, E. J.—UZU, F. O. (1972): Characteristics of phosphate adsorption by some Nigerian soils. *Soil Sci. Soc. Amer. Proc.*, **36**, 879–883.
- VIJ, A. C.—DEV, G. (1978): Availability of phosphorus to wheat in soils from North-West India as related to their phosphate adsorption. *J. Indian Soc. Soil Sci.*, **26**, 367–371.
- VIJAY CHANDRAN, V. K.—HARTAR, R. D. (1975): Evaluation of phosphorus adsorption by a cross-section of soil types. *Soil Sci.*, **119**, 119–126.

## DIALLEL ANALYSIS FOR SOME QUALITY TRAITS IN WHEAT (TRITICUM AESTIVUM L.)

It was considered worthwhile evaluating the important wheat varieties now being used extensively in the Indian wheat breeding programme with regard to their breeding value for some of the many quality criteria enumerated by AUSTIN—RAM (1971). The genetic analysis of 1000-grain weight, protein content and sedimentation value was attempted by ARORA—CHANDRA (1979). The present paper provides information on the genetic situation with regard to methionine content, Pelshenke value and phenol colour reaction.

Ten wheat varieties were chosen in view of their importance in the current wheat breeding programme, namely Hira, K-227, Sonora 64, Lerma Rojo, NP 809, PV 18, Sonalika, UP 301, Agra Local and C-306. A diallel set of crosses among these lines (excluding the reciprocals) was made. The resulting 45  $F_1$ s, along with the 10 parents, were planted in a randomized block design having 2 replications. Ten seeds of each entry were sown in 3 metre rows 30 cm apart with a plant-to-plant distance of 10 cm in each replication. Generally recommended doses of fertilizer and irrigation for this area were applied. Replication-wise samples were taken for each entry.

For the determination of methionine content, the method outlined by HORN *et al.* (1946) was used. The Pelshenke values were obtained following the method described by AUSTIN—RAM (1971). The phenol colour reaction of the polyphenol oxidase activity (tyrosinase activity) was assessed in accordance with ABROL *et al.* (1971). Data on methionine content and Pelshenke value were subjected to appropriate statistical analysis, i.e. analysis of variance for randomized block design to assess genotypic differences, followed by combining ability analysis according to Model I, method 2 of GRIFFING (1956), diallel analysis for components of variance and the graphic analysis of HAYMAN (1954).

The parents and hybrids indicated wide variation in mean values (Table 1) and significant genotypic differences (Table 2). The combining ability analysis (Table 3) revealed considerable non-additive genetic variance ( $\sigma^2_{sca}$ ) for these attributes in the presence of significant additive genetic variance ( $\sigma^2_{gca}$ ). The non-additive component for methionine content was about nine times the additive component, but for the Pelshenke value it was only twice the magnitude of the latter. By the analysis of HAYMAN (1954) the dominance component ( $H_1$ ) was observed to be about twice the value of the additive component (D) in respect of the Pelshenke value; however, it was nearly three times as great for methionine content (Table 4). Here again both estimates were statistically significant. It was noted that  $b_{wr}/v_r$  was non-significant, implying that much of the non-additive component, particularly the epistatic type of interaction, was important. The high estimate of mean degree of dominance (1.9) indicated overdominance, which again could be attributed at least partly to dominance and partly to non-allelic interaction. In the absence of significant regression  $b_{wr}/v_r$ , the  $H_1$  component tends to be overestimated and the degree of dominance obtained is generally a spurious indication of the dominance. The source of contribution to the observed epistasis was traced to varieties Hira, K-227 and C-306, characterised by negative values of  $W_r$ . The varieties Sonalika, UP 301 and Agra Local were identified as the best combining parents (Table 5)



for this character. The intercrosses of these three good combining parents exhibited high gca effects, which seemed to arise from the complementary action of additive genes. It was noted that the variety Hira, which is almost as high in mean methionine content as Sonalika, failed to give promising hybrids. Thus, high parental performance might not be simply related to high gca effects for methionine content. AHMED—MURTY (1972) studied gene action for methionine content in bajra and pointed out that the non-additive type of gene action governed methionine content.

**Table 1**

*Means of parents and  $F_1$  hybrids in a diallel cross of wheat for Pelshenke values (time in minutes, right of the diagonal) and methionine content (mg/g sample, left of the diagonal)*

Varieties	Hira	K-227	Sonora 64	Lerma Rojo	NP 809	PV 18	Sonalika	UP 301	Agra Local	C-306
Hira	<i>105.0</i> <i>2.90</i>	103.5	102.5	123.5	80.5	77.0	134.5	159.0	131.0	112.5
K-227	2.41	<i>88.5</i> <i>2.6</i>	110.0	108.5	62.0	68.0	93.5	105.5	65.5	50.5
Sonora 64	2.89	2.54	<i>150.0</i> <i>1.40</i>	125.5	76.0	130.0	153.5	176.5	123.5	59.0
Lerma Rojo	2.40	2.52	1.39	<i>78.5</i> <i>1.40</i>	85.0	67.0	104.0	101.0	100.0	105.0
NP 809	1.84	1.40	1.17	2.54	<i>58.5</i> <i>2.54</i>	69.0	62.0	111.5	88.0	99.5
PV 18	2.53	1.10	1.60	2.26	2.72	<i>102.5</i> <i>1.40</i>	68.5	168.5	124.5	90.5
Sonalika	1.90	1.16	1.36	2.03	2.96	2.91	<i>69.5</i> <i>2.91</i>	131.0	84.5	58.5
UP 301	1.12	1.60	2.07	2.54	3.00	2.91	3.00	<i>140.0</i> <i>2.40</i>	132.5	87.0
Agra Local	2.97	1.50	1.98	1.50	2.95	2.91	2.94	2.99	<i>90.0</i> <i>2.20</i>	96.5
C-306	1.28	2.95	2.26	2.23	2.54	2.24	2.82	2.54	2.21	<i>98.5</i> <i>1.83</i>

Figures in italics indicate parental values

**Table 2**

*Analysis of variance for randomized block design in respect of methionine content and Pelshenke value in a diallel cross of wheat*

Source	d.f.	Mean square	
		Methionine content	Pelshenke values
Replications	1	0.002	33.8277
Progenies	54	0.977**	1802.6839**
Error	54	0.001	30.4570

\*\* Significant at 1% level of probability.

**Table 3**

*Analysis of variance for combining ability in a diallel cross of wheat with regard to methionine content and Pelshenke value*

Source	d.f.	Mean squares	
		Methionine content	Pelshenke values
General combining ability (gca)	9	0.595**	1978.329**
Specific combining ability (sca)	45	0.374**	485.943**
Error	54	0.0005	75.228
$\sigma^2$ gca		0.495	241.919
$\sigma^2$ sca		0.3735	410.715

\*\* Significant at 1% level of probability.

**Table 4**

*Estimates of regression slope (vr, wr) and genetic components of variance with regard to methionine content and Pelshenke value in a diallel cross of wheat*

Estimate	Methionine content	Pelshenke values
$^b(wr, vr)$	0.51 $\pm 0.15$	0.56 $\pm 0.16$
D	0.41 $\pm 0.12$	811.93 $\pm 172.10$
H <sub>1</sub>	1.59 $\pm 0.26$	1931.81 $\pm 336.32$
H <sub>2</sub>	1.37 $\pm 0.22$	1791.12 $\pm 311.33$
h <sup>2</sup>	0.80 $\pm 0.14$	6.59 $\pm 208.39$
F	-0.45 $\pm 0.14$	-135.59 $\pm 297.08$
Mean degree of dominance (H <sub>1</sub> /D) <sup>1</sup>	1.97	1.54
Proportion of genes with +ve and -ve effects (H <sub>2</sub> /4 H <sub>1</sub> )	0.21	0.24
Proportion of dominant and recessive genes in parents $\frac{(4 DH_1)^{1/2} + F}{(4 DH_1)^{1/2} - F}$	1.77	0.89
Correlation r (Wr + Vr), Yr	0.23	0.49
Number of dominant genes for each recessive gene $\frac{h^2}{H_2}$	0.58	0.003

The range of parental means for the Pelschenke value was reasonably high, i.e. between 58.5 and 150 (Table 1). The estimates of mean squares due to gca and sca were both highly significant (Table 3). Mainly non-additive gene action appeared to be operative in the material, but the additive component was also significant in the expression of the character. From the Hayman's analysis of the components of variance, it was found that the additive portion, D, was about half the dominance component,  $H_1$ . Also, the regression coefficient  $b_{wv}/v_r$  turned out to be significantly different from unity, indicating a departure from assumptions of diallel analysis attributable primarily to non-allelic interaction. The parent C 306 was perhaps responsible for this, as it had a negative value of  $W_r$ . AUSTIN—RAM (1971) worked out the

Table 5

*Estimates of gca effects with regard to methionine content and Pelschenke value among parents of a diallel cross in wheat*

No.	Parents	Methionine content	Pelschenke test values
1.	Hira	0.08	10.39
2.	K 227	-0.37	-13.77
3.	Sonora 64	-0.34	20.64
4.	Lerma Rojo	0.18	-2.73
5.	NP 809	0.17	-21.57
6.	PV 18	-0.01	-3.44
7.	Sonalika	0.23	-6.69
8.	UP 301	0.20	28.56
9.	Agra Local	0.18	1.39
10.	C-306	0.04	-12.77
SE(gi)		0.002	1.06
SEd(gi-gj)		0.004	1.59
CD (p = 0.05)		0.007	3.15

Table 6

*Phenol colour reaction scores in parents and  $F_1$  hybrids of diallel cross in wheat*

Varieties	Hira	K-227	Sonora 64	Lerma Rojo	NP 809	PV 18	Sonalika	UP 301	Agra Local	C-306
Hira	2	3	2	3	3	3	3	2	3	3
K-227		4	2	2	3	3	4	4	3	4
Sonora 64			2	2	2	3	3	1	3	1
Lerma Rojo				2	2	1	3	2	2	3
NP 809					2	3	3	2	2	2
PV 18						2	4	2	3	3
Sonalika							4	2	4	4
UP 301								2	4	2
Agra Local									4	4
C-306										4

Figures indicate parental values; 1 — deep brown; 2 — light brown; 3 — pink; 4 — colourless.



Pelshenke values of various wheat varieties and found that the strength of the gluten was helpful in defining gluten or dough strength or both in relation to chapati making quality.

A perusal of the data on polyphenol oxidase activity (tyrosinase activity, Table 6) revealed a trend of additive gene action. However, some of the  $F_1$  hybrids also behaved in favour of a simple dominance mechanism. Only a few hybrids showed interaction of the over-dominance type. However, these data were not subjected to any statistical analysis and it was not possible to predict from the present data how this would behave in progenies of the hybrids. The desirable parents for polyphenol oxidase activity were identified as Sonalika and K 227. This test not only constitutes a simple and swift indicator of chapati quality in wheat but also appears to be a relatively simply inherited trait (ABROL *et al.* 1971). Therefore, a careful study of the inheritance of this trait is warranted.

A consideration of the mean performance of individual hybrids, their sca effects, as well as the gca effects of the parents involved, revealed that hybrids involving Sonora 64 and Sonalika as parents figured among the best performing ones. The best hybrid for methionine content was Sonalika  $\times$  UP 301, while Sonora 64  $\times$  UP 301 was the best for Pelshenke value. It should be possible to attain high and significant gains simultaneously for the different quality characters if the best crosses are recombined by first making double crosses between any two of them at a time and then attempting a multiple cross among the best selected plants in the progenies of double crosses. This programme has been taken up in the quality improvement project for wheat.

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### References

- ABROL, Y. P.—UPRETI, D. C.—RAM, A.—TIKKO, S. (1971): Phenol colour reaction is an indicator of chapati quality in wheat. *Sabao News Letter*, 3/1, 17–21.
- AHMED, Z.—MURTY, B. R. (1972): Inheritance of protein and essential amino acids in  $22 \times 22$  partial diallel of *P. typhoides*. *Indian J. Genet.*, 32, 400–407.
- ARORA, S. K.—CHANDRA, S. (1979): Genetic analysis of some quality characters in wheat. *Indian J. Genet.*, 39, 67–76.
- AUSTIN, A.—RAM, A. (1971): Studies on chapati making quality of wheat. I.C.A.R., New Delhi, *Tech. Bull.*, 31.
- GRIFFING, B. (1956): Concept of general and specific combining ability in relation to diallel crossing system. *Aust. J. Biol. Sci.*, 9, 463–493.
- HAYMAN, B. I. (1954): The theory and analysis of a diallel cross. *Genetics*, 45/1, 155–172.
- HORN, M. J.—JONES, D. B.—BULM, A. E. (1946): The estimation of methionine content. *J. Biol. Chem.*, 166, 313–320.

### SEEDLING HANDEDNESS IN TRITICALE AND ITS PARENTS

Seedling handedness in *Secale cereale* L. was first observed by COMPTON (1912), who remarked that “*Secale cereale* is an unfavourable plant for this purpose, owing to the narrowness of the leaves and the frequency with which both margins are curved inwards in the upper portion. Out of 30 seedlings 16 were left-handed and 14 were right-handed, showing that both conditions occur here also, though the numbers are insufficient to allow a ratio to be calculated.”

Table 1

*Review of seedling handedness in the tribe Triticeae (Gramineae)*

No.	Genus/Species	References
1.	<i>Triticum monococcum</i>	Einkorn (Diploid) ONO—SUEMOTO 1957
2.	<i>T. aegilopoides</i>	Wild Einkorn (Diploid) ONO—SUEMOTO 1957
3.	<i>T. durum</i>	Emmer (Tetraploid) ONO—SUEMOTO 1957
4.	<i>T. turgidum</i>	Conewheat (Tetraploid) ONO—SUEMOTO 1957
5.	<i>T. timopheevi</i>	Timopheevi (Tetraploid) ONO—SUEMOTO 1957
6.	<i>T. dicoccoides</i>	Wild Emmer (Tetraploid) KIHARA 1972
7.	<i>T. dicoccum</i>	Emmer (Tetraploid) KIHARA 1972
8.	<i>T. persicum</i>	Persian (Tetraploid) KIHARA 1972
9.	<i>T. aestivum</i>	Dinkel (Hexaploid) ONO—SUEMOTO 1957
10.	<i>T. spelta</i>	Spelta (Hexaploid) ONO—SUEMOTO 1957
11.	<i>T. sphaerococcum</i>	Indian swarf wheat (Hexaploid) KIHARA 1972
12.	<i>T. macha</i>	Macha Wheat (Hexaploid) KIHARA 1972
13.	<i>T. pyramidale</i>	— KIHARA 1972
14.	<i>Aegilops squarrosa</i>	— KIHARA 1972
15.	<i>Secale cereale</i>	— COMPTON 1912
16.	<i>Hordeum distichum</i>	— COMPTON 1912
17.	<i>H. hexastichum</i>	— COMPTON 1912

Seedling handedness in the genus *Triticum* was first observed by ONO—SUEMOTO (1957). KIHARA (1972) reviewed the subject and dealt with various aspects of handedness in *Triticum* in detail. In Table 1, a review of seedling handedness in the tribe Triticeae is given.

Historically  $\times$ Triticosecale Wittmack or *Triticale* is the intergeneric hybrid between *Secale  $\times$  *Triticum* and both belong to the tribe Triticeae (*Gramineae*). GUSTAFSON (1976) has reviewed the evolutionary development of *Triticale* but makes no comment on seedling handedness. CHOUDHARY—NIRMALA (1976) studied the effects of gamma-rays and P-fluorophenyl-alanine on seedling heights and chromosome aberrations in *Triticale*, but failed to observe seedling handedness.*

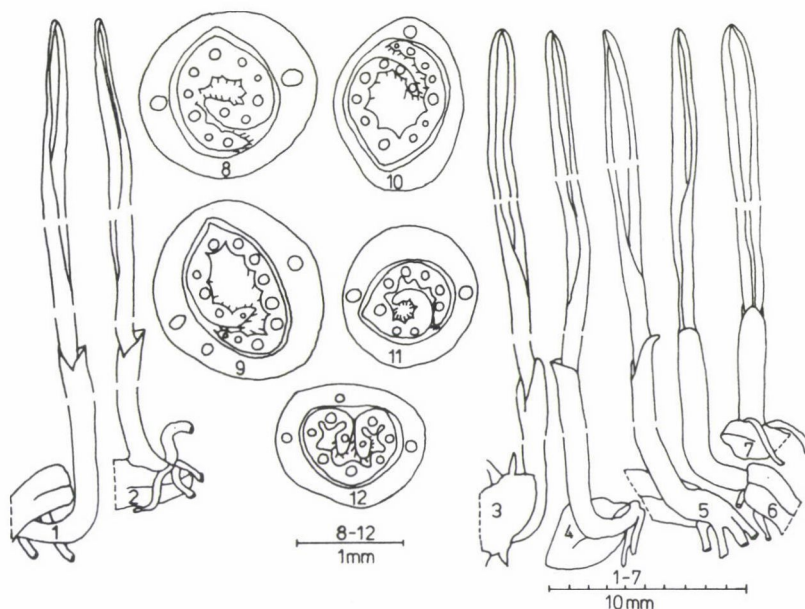
In this communication preliminary observations on seedling handedness are described in  $\times$ *Triticale*, which is hitherto unknown, together with seedling handedness in its parents, viz. *Triticum* and *Secale*.

For the study of handedness seed material of *Triticale* and its parents were sown separately in petri dishes containing moistened blotters and periodically examined. Handedness was detected in the seedling stage, when the first leaf above the coleoptile begins to unfold 4–6 days after sowing. Handedness can be easily detected in young seedlings by either dissecting it or cutting it transversely (Figs 1–12). Depending on the folding of the first leaf either to the left or right, a seedling may be classified as left-handed or right-handed. Occasionally there is no folding of the first leaf and such seedlings are designated as neutral (Figs 7 and 12).

The ratio of left- and right-handed seedlings and neutral seedlings in *Triticale* and its parents were scored separately and analysed statistically.

In Table 2, data on seedling handedness in *Triticale* and its parents are summarised. A total of 2000 *Triticale* seedlings were examined, out of which 1055 were left-handed (52.75%) and 913 were right-handed (45.65%). The  $\chi^2$  for deviation for left- and right-handed seedlings





Figs 1-2. Right- and left-handed seedlings of *Triticale* (DTS-642)

Figs 3-4. Right- and left-handed seedlings of *Triticum* (UP-215)

Figs 5-7. Right- and left-handed seedlings and neutral seedlings of *Secale cereale*

Figs 8-9. Transverse section of right- and left-handed seedling across the coleoptile, showing the folding of the leaf to the right or left, respectively

Figs 10-12. Transverse section of right-, left-handed and neutral seedlings across the coleoptile, showing the folding of the leaf to the right or left. In the neutral seedling, the margins of the leaf meet but do not fold

is high ( $\chi^2 = 10.594$ ) and the P-value is less than 1%, which is highly significant. In all but one (DTS-642) of the varieties of *Triticale* examined there were significantly more left-handed seedlings than right-handed ones. Similarly, the L/R ratio in most of the varieties is equal to unity, whereas in DTS-642 it is less and in DTS-281-4 and 216-8 it is greater than unity. The  $\chi^2$  values of DTS-281-4 and 216-8 show significant P-values (Table 2).

In *Triticum aestivum*, a total of 1800 seedlings were examined, out of which 902 were left-handed (50.11%) and 873 were right-handed (48.5%). The  $\chi^2$  value is 0.814 and the P-value for 1 df is not significant. In all the *Triticum* varieties (Table 2) there is an excess of left-handed seedlings over right-handed ones. But in NI-5439 and UP-215 there were found to be more right-handed seedlings than left-handed ones, though the deviation from equality was not significant. The L/R ratio in all the varieties studied is unity, however (Table 2).

In *Secale cereale* a total of 1443 seedlings were scored, but strangely only 19 seedlings were found to be right-handed (1.346%), while 12 were left-handed (0.849%) and the P-values were not statistically significant (Table 2).

In *Triticale* the number of neutral seedlings examined was 32 out of a total of 2000, i.e. 1.6%, and in *Triticum* there were 25 neutral seedlings (1.389%) out of a total 1800 scored, whereas in *Secale cereale* a total of 1412 neutrals were observed out of a total of 1443 seedlings scored. The percentage of neutrals in *Secale cereale* is thus very high (97.08%) in comparison to that of *Triticale* and *Triticum* (Table 2). Thus, *Secale cereale* is probably more primitive than *Triticale* and *Triticum* with regard to the folding of the first leaf.



Table 2

*Ratios of left- and right-handed seedlings in Triticale and its parents*

Species/ Variety	Number of seedlings scored	Left	Right	L - R	L + R	L/R	N	$\chi^2$ for 1 : 1 deviation for L and R	P value, %
<i>Triticale</i>									
DTS-42-3	200	101	89	12	190	1.134	10	1.22	30
DTS-642	300	137	158	-21	295	0.867	5	1.553	20
DTS-47-1	300	154	139	15	293	1.110	7	0.913	30
DTS-280-7	300	156	142	14	298	1.0982	2	0.807	30
DTS-205-4-1	300	155	142	13	297	1.090	3	0.593	50
DTS-281-4	300	169	128	41	297	1.320	3	5.633	5
DTS-216-8	300	183	115	68	298	1.590	2	15.43	1
Total	2000	1055	913	142	1968	1.15	32	10.594	1
<i>Triticum</i>									
NI-747-19	300	152	144	8	296	1.056	4	0.267	70
NI-5439	300	145	151	-6	296	0.60	4	0.173	70
Sonalika	300	153	142	11	295	1.077	5	0.487	50
Kalyanasona	300	151	142	9	293	1.063	7	0.433	50
UP-215	300	146	149	-3	295	0.980	5	0.113	70
HD-1739	300	155	145	10	300	1.062	—	0.333	50
Total	1800	902	873	29	1775	1.033	25	0.814	30
<i>Secale cereale</i>	1443	12	19	-7	31	0.6366	1412	1.581	20

% of neutrals in *Triticale* = 1.6, % of neutrals in *Triticum* = 1.389 and % of neutrals in *Secale cereale* = 97.80

A comparison of the data on seedling handedness with the existing data on *Hordeum distichum* and *H. hexastichum* showed a great excess of left-handers (COMPTON 1912). The L/R ratio in these species is 1.397 and 1.282, respectively. Similarly in *Secale cereale* an excess of left-handed seedlings (by two) was observed in a small population of 30 plants (COMPTON 1912). The *Triticale* and *Triticum* presently studied also showed an excess of left-handed seedlings. A similar excess of left-handed seedlings was observed in *Setaria italica* (COMPTON 1912), *Pennisetum americanum*, *Sorghum vulgare* (BAHADUR—UDAYACHANDRA 1980) and in several genera and species of *Gramineae* (BAHADUR *et al.* 1980). In *Bambusa arundinacea* and *Avena sativa* an excess of right-handed seedlings was observed by BAHADUR *et al.* (1978) and by COMPTON (1912), respectively. It is interesting to remark that the ratio of left- and right-handedness in plants or plant organs in different species, although unrelated, has been found to be similar to the one presently studied (DAVIS 1974, DAVIS—RAMANUJACHARYULU 1971, BAHADUR *et al.* 1979). Thus, seedling handedness in *Triticale* and its parents represents a case of bioisomerism and this is correlated with mathematical isomorphism (MAYEN 1973), as evidence by the equality of the L/R ratio.

COMPTON (1912) states, "There appears to be conclusive evidence that the direction of folding of the first leaf is not inherited" and further states "the L.H./R.H. is hereditary, though right and left handedness themselves are not", and concludes that the difference in the shape of the environment and the developing embryo appears to be the probable cause

of handedness. It is of interest to remark that DAVIS (1974) also regards handedness in coconut palms as not genetical and considers handedness in coconuts to be due to geophysical influence, i.e. relationship between latitude and handedness.

It is thus obvious that the seedling handedness in Triticale and its parents presently studied represents a mirror image pattern or stereoisometric phenomenon (DORMER 1965). In the recent literature handedness in plants or plant organs has been referred to as isomerism (BAHADUR—VENKATESHWARLU 1976a, b), bioisomerism (BAHADUR *et al.* 1978, 1979) and enantiomorphic structures (DAVIS 1974). Recently, BAHADUR *et al.* (1979) have proposed a classification of plant handedness and have discussed the various aspects of this phenomenon. MCGAVIN (1976) has similarly proposed a classification of chiral biological structures, and under category II includes developmental handedness, showing enantiomorphism comparable to that presently studied.

Although there is no clear evidence to explain the nature of the handedness studied here, it is probable that the handedness is due to molecular chirality, which is inherent in the organisms and is expressed in the shape of biological asymmetry (chirality) during morphogenesis (BAHADUR *et al.* 1979). Further work on various aspects of this interesting phenomenon will be published in due course.

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#### References

- BAHADUR, B.—VENKATESHWARLU, T. (1976a): Isomerism in flowers of *Carica papaya* L. J. Indian Bot. Soc., **55**, 30–37.
- BAHADUR, B.—VENKATESHWARLU, T. (1976b): Isomerism in flowers of four species of *Jatropha* L. J. Indian Bot. Soc., **55**, 89–94.
- BAHADUR, B.—RAO, K. L.—RAO, M. M. (1978): The left and right handedness of seedlings in *Bambusa arundinacea*. Curr. Sci., **47**, 584–586.
- BAHADUR, B.—RAO, K. L.—KUMAR, P. V.—REDDY, N. P.—RAO, M. M. (1979): Some aspects of handedness in plants. In: Vistas in molecular soil state and biophysics. Prof. P. G. Puranic 60th Birthday Commemorative Volume, Hyderabad, 355–363.
- BAHADUR, B.—UDAYACHANDRA, U. S. (1980): The right and left handedness of seedlings in *Sorghum*. Indian J. Bot., **3**, 116–121.
- BAHADUR, B.—RAMASWAMY, N.—REDDY, M. M. (1980): Some aspects of seedling handedness in Gramineae. In: Natl. Symp. Life Sci., Nagpur (Abstract No. 282).
- CHOUDHARY, J. B.—NIRMALA (1976): Effect of Gamma-rays on seedling height and chromosome aberrations in Triticale. Acta Bot. Indica, **4**, 126–130.
- COMPTON, R. N. (1912): A further contribution to the study of right and left handedness. J. Genet., **2**, 53–70.
- DAVIS, T. A. (1974): Enantiomorphic structures in plants. Proc. Indian Natl. Sci. Acad. B., **40**, 424–429.



- DAVIS, T. A.—RAMANUJACHARYULU, C. (1971): Statistical analysis of bilateral symmetry in plant organs. *Sankhya, Indian J. Stat. B.*, **33**, 259–290.
- DORMER, K. J. (1965): Correlations in plant developments. General and basic aspects. In: *Encyclopedia of plant physiology*, Ed. W. Ruhland. Springer Verlag. New York. XV/1, 452–478.
- GUSTAFSON, J. P. (1976): The evolutionary development of Triticale: The wheat rye hybrid. In: *Evolutionary Biology*, **9**, 107–135.
- KIHARA, H. (1972): Right and left handedness in plants — A review. *Seiken Zihō*, **23**, 1–37.
- MAYEN, S. V. (1973): Plant morphology in its nomothetical aspects. *Bot. Rev.*, **39**, 205–260.
- MCGAVIN, S. (1976): The handedness or chirality of biological structure at the molecular and at higher levels of structural organisations. *Biosystems*, **8**, 147–152.
- ONO, H.—SUEMOTO, H. (1957): The right and left handedness of seedlings in *Triticum*. *Seiken Zihō*, **8**, 60–66.

### COMPARATIVE STUDY ON HYBRID MAIZES OF DIFFERENT MATURITY TIME AND GENETIC STRUCTURE

Of the grain fodder requirements of Hungary, some 70 per cent is covered by maize, which also ensures about 30 per cent of the protein supply of the livestock. Beside its high energy content, the maize contains relatively little of protein, and even this protein — which mostly consists of zein — is of unfavourable amino acid composition. The amount of lysine, an amino acid indispensable for swine, is particularly low in the maize protein. That is why, in Hungary too, attempts are made to introduce and spread the opaque varieties besides the normal hybrids, since the former contain 60–80 per cent more lysine than the latter, due to the changed proportion of zein in the endosperm of the grain (NOTHEISZ *et al.* 1977). In addition, the opaque maize contains some 25 per cent more fat than the normal hybrids.

In experiments with rats, hybrids with normal endosperm were compared with opaque and waxy hybrids to find out their biological value, while their nutrient content as well as lysine and available lysine contents were established by chemical analysis.

The experiments included 7 normal, 6 opaque and 1 waxy maize hybrids of various maturity time, obtained from the trial grounds of the National Institute for Agricultural Variety Trial.

The harvested grain crop of the 14 maize hybrids was dried in laboratory thermostat at 60 °C. Their nutrient contents (dry matter-, crude protein-, crude fat-, raw fibre-, crude ash- and N-free extractable matter content) were determined according to the standard MSZ 6830.T./1965.

The lysine contents of the maize samples were determined by a BC 200. type automatic amino acid analyser (according to the instructions of the manual published by the BIO-GAL company in 1971), while the available lysine content was measured on the basis of CARPENTER's (1960, 1973) method.

Nitrogen turnover experiments were carried out with male albino rats, of an average weight of 80–90 g, each kept in a metabolism cage. On the basis of the data of N-turnover tests, we determined the biological value, net conversion, apparent digestibility and productive conversion of proteins contained in the maize samples (SZELÉNYI 1969). Having completed the N-turnover tests, we killed the rats and determined their dry matter, crude protein and crude fat contents.

In Tables 1 and 2 the crude protein, total lysine and available lysine as well as crude fat contents, further the yields of normal, opaque and waxy maizes are grouped according to the time of maturing. The dry matter content of the maize samples was 90–91 per cent; crude protein and crude fat contents are given as a percentage of dry matter. Maizes with normal endosperm mostly had crude protein contents between 10.3 and 10.5 per cent; only



Mv SC 580 contained 9.0 and Szegedi SC 369 9.3 per cent crude protein. The highest crude fat content was found in SC 1584 (4.9 per cent) and Pioneer 3709 (4.7 per cent). In the other maize samples, the crude fat content ranged from 3.9 to 4.1 per cent. In the normal hybrids, the total lysine content was 0.23–0.25 per cent, while the available lysine content was found to be the highest in Szegedi SC 369 (82.2 per cent) and the lowest in Mv SC 580 (76.5 per cent) (Table 1).

**Table 1**  
*Yield and nutrient content of maize hybrids with normal endosperm*

	Yield, t/ha	Crude protein content	Total lysine in dry matter	Available lysine content	Crude fat content
				%	
1. Pioneer 3965 A MTC FAO 200	9.62	10.4	0.23	81.6	4.0
2. Szegedi SC 369 FAO 300	8.40	9.3	0.24	82.2	4.1
3. JX 92 SC FAO 300	9.93	10.5	0.25	78.7	4.8
4. Szegedi MSC 378 FAO 300	10.45	10.3	0.24	80.0	4.0
5. Pioneer 3709 MSC FAO 400	11.02	10.4	0.25	80.5	4.7
6. Mv SC 580 FAO 500	8.20	9.0	0.23	76.5	3.9
7. SC 1584 FAO 500	10.66	10.4	0.25	79.3	4.9

**Table 2**  
*Yield and nutrient content of opaque and waxy maize hybrids*

	Yield, t/ha	Crude protein content	Total lysine in dry matter	Available lysine content	Crude fat content
			%		
8. SC 3365 opaque FAO 400	7.20	9.8	0.34	83.2	4.7
9. SC 3385 opaque FAO 400	7.92	9.6	0.32	84.7	5.0
10. SC 5443 opaque FAO 400	8.25	8.9	0.32	82.7	5.1
11. SC 5553 opaque FAO 500	7.86	8.5	0.27	76.3	4.5
12. TC 3560 opaque FAO 500	8.18	8.2	0.25	75.9	4.8
13. MO 2530 opaque FAO 500	5.80	9.9	0.27	77.0	4.5
14. SC 1554 waxy FAO 500	10.25	10.2	0.24	78.4	4.6

The crude protein content generally was lower in the opaque than in the normal hybrids. The highest crude protein content (9.9 per cent) was measured in MO 2530, while the lowest (8.2 per cent) in TC 3560. The waxy hybrid contained 10.2 per cent crude protein. The crude fat contents of opaque maizes ranged from 4.5 to 5.1 per cent, while that of the waxy hybrid was 4.6 per cent. The highest total lysine contents were measured in SC 3365 (0.34 per cent) and SC 3385 (0.32 per cent) while the quantities of total lysine found in SC 1553, MO 2530 and TC 3560 were similar to those of the normal maizes (0.25–0.27 per cent). The available lysine content was 82.7–84.7 per cent in the medium late and 75.9–77.0 per cent in the late opaque hybrids.

The yields of maize hybrids with normal endosperm — except Szegedi SC 369 and Mv SC 580 obtained from the experimental production of opaque maize — ranged between 9.65 and 11.02 ton/ha. The results suggest a positive correlation between vegetation period and productivity. Under the conditions of Hungary Pioneer 3709 MSC, a maize hybrid with excellent production potential and stalk strength can be reliably grown on 70 per cent of the sowing area of the country. Szegedi MSC 378 gives a similarly favourable yield; however, owing to the low cold tolerance of its maternal component, its cultivation requires increased care. Pioneer 3965 A MTC deserves attention because of its outstanding productivity, stalk strength and quick release of water.

Some opaque hybrids (SC 5443, TC 3560) produced yields close to that of Szegedi SC 369 (8.4 ton/ha), the maize included in the experiment as a standard, and were found to be nearly identical with it in other qualities too. Nevertheless, the economically disadvantageous characteristics of the opaque hybrids (e.g. release of water, susceptibility of ears to fusarium

**Table 3**  
*Protein conversion indices of normal maize hybrids*

	Weight increase	Nitrogen balance	BV	NPC	APD	PPC
	of rats					
	g/day	mg/day				
1. Pioneer 3965 A MTC FAO 200	1.2	60	68 ± 2.6	62 ± 3.2	77 ± 2.3	33 ± 2.0
2. Szegedi SC 369 FAO 300	1.2	58	74 ± 5.7	71 ± 5.0	80 ± 2.3	38 ± 5.4
3. JX 92 SC FAO 300	1.2	63	73 ± 4.8	66 ± 3.6	81 ± 6.2	38 ± 6.7
4. Szegedi MSC 378 FAO 300	1.6	72	75 ± 2.5	69 ± 3.7	79 ± 3.2	41 ± 3.2
5. Pioneer 3709 MSC FAO 400	1.6	76	74 ± 2.7	67 ± 2.9	79 ± 2.2	41 ± 3.7
6. Mv SC 580 FAO 500	1.0	42	58 ± 2.8	55 ± 2.9	81 ± 1.1	25 ± 3.0
7. SC 1584 FAO 500	1.4	66	67 ± 4.4	63 ± 4.9	80 ± 3.0	34 ± 5.4

Abbreviations: BV = biological value  
NPC = net protein conversion  
APD = apparent protein digestibility  
PPC = productive protein conversion

etc.), which in cool, rainy years may have an unfavourable effect on productivity and reliability of production, must not be left out of consideration.

In a nitrogen turnover experiment carried out with white rats, the changes of body weight and the daily N balance were determined in a five-day experimental phase following four days of pre-feeding. Of the normal hybrids, Pioneer 3709 MSC and Szegedi MSC 378 caused the highest daily weight increase (1.6 g/day). With the other maizes fed to the animals, a weight increase of 1.2–1.4 g/day was obtained, except the hybrid Mv SC 580 which resulted in a mere 1.0 g increase of weight a day (Table 3). At the same time, animals consuming the medium late opaque maize exhibited a daily weight increase of 2.0–2.2 g, while with the late opaque hybrids only a 0.8–1.0 g/day increase of weight was achieved. Increase in body weight, checked after feeding the waxy maize, was also relatively low — 1.2 g a day (Table 4).

The lowest daily N retention was obtained with Mv SC 580 (42 mg) and Szegedi SC 369 (58 mg). Exceedingly high N retention was observed after the consumption of Pioneer 3709 MSC (76 mg/day) and Szegedi MSC 378 (72 mg/day) (Table 3).

In the course of feeding the opaque maizes SC 3365, SC 3385 and SC 5443, the daily N balance was 78–85 mg; while with SC 5533 and TC 3560, it was 21–24 mg, and in the case of the waxy hybrid 63 mg (Table 4).

The biological value of Szegedi SC 369, Szegedi MSC 378, JX 92 SC and Pioneer 3709 MSC was 73–75 per cent, higher than that generally found in normal hybrids. The 67–68 per cent biological value of SC 1584 and Pioneer 3965 MTC proves the high quality of protein; while Mv SC 580 exhibits the biological value of a medium quality hybrid maize (58 per cent).

The data of net protein conversion — which express how much of the protein consumed has been utilized — show a similar tendency. The values of apparent protein digestibility

**Table 4**  
*Protein conversion indices of opaque and waxy maize hybrids*

	Weight increase	Nitrogen balance	BV	NPC	APD	PPC
	of rats					
	g/day	mg/day				
8. SC 3365 O <sub>2</sub> FAO 400	2.0	85	78 ± 3.6	78 ± 3.2	86 ± 0.8	49 ± 3.4
9. SC 3385 O <sub>2</sub> FAO 400	2.2	78	76 ± 2.2	73 ± 2.0	81 ± 0.9	43 ± 1.4
10. SC 5443 O <sub>2</sub> FAO 400	2.0	80	85 ± 2.5	83 ± 1.7	82 ± 3.9	52 ± 1.8
11. SC 5533 O <sub>2</sub> FAO 500	1.0	21	47 ± 1.8	44 ± 1.8	77 ± 2.8	10 ± 1.3
12. TC 3560 O <sub>2</sub> FAO 500	0.8	24	54 ± 3.6	49 ± 3.8	74 ± 4.0	13 ± 4.1
13. MO 2530 O <sub>2</sub> FAO 500	0.8	57	62 ± 2.7	58 ± 1.9	79 ± 2.2	29 ± 1.6
14. SC 1554 waxy FAO 500	1.2	63	67 ± 4.4	63 ± 4.9	80 ± 3.0	34 ± 5.4

Abbreviations: BV = biological value  
NPC = net protein conversion  
APD = apparent protein digestibility  
PPC = productive protein conversion



ranged from 79 to 81 per cent; the lowest of them (77 per cent) was that of Pioneer 3965 MTC. The productive protein conversion — which expresses the relation between N balance and consumed N — was the best with maizes showing the highest biological value, and the poorest (25 per cent) within the normal hybrid Mv SC 580 (Table 3).

Extremely high biological value (85 per cent) was obtained with the opaque maize SC 5443, and the hybrids SC 3365 and SC 3385 also showed good biological values (76–78 per cent). The biological value was lower than expected with MO 2530 (62 per cent), SC 5533 (47 per cent) and TC 3560 (54 per cent). The 67 per cent biological value of the waxy maize gives evidence of a good quality protein.

The net protein conversion is — in the previous order again — excellent with the opaque hybrids SC 5443 (83 per cent), SC 3385 (73 per cent) and SC 3365 (78 per cent). It is the poorest with SC 5533 (44 per cent) and TC 3560 (49 per cent). The 86 per cent value of apparent protein digestibility with SC 3365 is outstanding, though the waxy hybrid SC 1554 with its 80 per cent, and the hybrids SC 3385 and SC 5443 with 81 and 82 per cent, respectively, were also found good. The value of apparent protein digestibility was the lowest (74 per cent) in the case of TC 3560.

The highest value of productive protein conversion (52 per cent) was obtained with the hybrid SC 5443; the opaque maizes SC 3365 and SC 3385 were also good in that respect (49 and 43 per cent, respectively). The 29 per cent productive protein conversion of MO 2530 suggests a medium protein content, while with SC 5533 and TC 3560 the value of productive protein conversion is very low (10 and 13 per cent, respectively). The 34 per cent productive protein conversion of the waxy hybrid SC 1554 is considered good (Table 4).

The bodies of rats killed after the completion of the N-turnover tests contained 27–30 per cent dry matter; the crude protein content ranged between 14.8 and 17.0 per cent and the crude fat content between 5.2 and 7.7 per cent. According to the general experience, the consumption of higher biological value opaque or normal maize hybrids resulted in an increased fat content of the body. (The coefficient of correlation between body fat content and biological value is 0.45.)

Of the maizes examined, those accepted for commercial production had a higher than 10 per cent crude protein content in the dry matter, on the average. This is much more than the crude protein content of maizes grown in Hungary in general. In the opaque maizes, on the other hand, the crude protein content was — with a few exceptions — lower than expected.

Maizes with normal endosperm contained 0.23–0.25 per cent lysine in the dry matter, a value corresponding to the Hungarian average. Foreign authors (CROMWELL *et al.* 1967, ROSA *et al.* 1977, RIVERA *et al.* 1978) pointed out 0.23–0.38 per cent lysine in the dry matter of normal hybrids, while KLEIN *et al.* (1971) determined 2.9–3.0 per cent lysine in the protein.

At the same time the lysine content of opaque maizes is given as 0.41% by ROSA (1977), 0.35% by PICK (1971), 0.38% by WAHLSTROM (1977), 0.49% by CROMWELL (1967), 0.41% by ANASTASIJEVIC (1978) and 0.40–0.46% by RIVERA (1978). KLEIN (1971) found 3.1–4.9 per cent lysine in the protein of opaque maizes. In the course of comparative studies on six different opaque maize hybrids, EGGUM (1979) pointed out great differences in lysine content; namely, the total lysine content of protein ranged from 3.54 to 4.18 per cent in his experiments. HARPSTEAD (1971) reports 3.39 per cent lysine content for the opaque and 2.0 per cent for the normal hybrids. From the data of the above listed authors and our own investigations, we have drawn the conclusion that the lysine content of the opaque hybrids may be highly varying.

Of the chemically demonstrated total lysine content, it is only the amount of available lysine that the animal is able to utilize. That is why we thought it very important to determine it; the more so because we searched for and did find a correlation between the biological value

calculated on the basis of N-turnover tests and the available lysine content. The coefficient of correlation is 0.84. As seen from our data, the available lysine content was always essentially higher in the early and medium late normal and opaque hybrids than in the late ones.

The biological value of early and medium late normal hybrids, though generally good, does not, naturally, reach the level attained with the good quality opaque hybrids. It can be established, on the other hand, that the biological value of proteins in late opaque and normal hybrids (FAO 500) is significantly lower.

The biological value of the waxy maize was only equal to that of the normal hybrids, but its high amylopectine content (99 per cent) is very favourably utilized in the animal organism (ROSA 1977). We note here that, in the normal hybrids, 70 per cent amylopectine and 30 per cent amylase were found.

It is worth mentioning that, when examining in another experiment series the conversion of proteins of various extracted soya meals in rats, we obtained values similar to those of proteins of good quality (early and medium late) opaque hybrids. With the best soya samples an 82–88 per cent biological value and a 52–57 per cent productive conversion were achieved at a 2.2 g daily rate of weight increase.

On the basis of our comparative studies, we suggest to encourage the introduction of medium late opaque maizes in commercial production; though at present the maize farms show reluctance to grow opaque hybrids because of the about 8–10 per cent smaller yield they produce. The quantitative view still prevalent in maize production must by all means be replaced by a demand for quality. We note here that, while the amount of lysine obtained with normal maize hybrids is some 15–18 kg/ha, the opaque hybrids produce 22–23 kg/ha lysine. In addition, the fact that the opaque hybrids contain about 25 per cent more fat than the normal ones must not be left out of consideration.

Furthermore, according to the results of examinations, the proteins of the normal hybrids, but first of all of the high lysine content opaque maizes, if they have a long vegetation period, are very poorly utilized in the animal organism, even if they occasionally contain more lysine.

The examinations unequivocally show that the vegetation period and the available lysine content are in negative correlation. Further, the feeding value of hybrids greatly depends on the grain/germ ratio, since it is in the germ that a large proportion of essential amino acids, determining the quality of protein, is localized. For this very reason, not only the grain/germ ratio but the thousand-grain-weight and through it the total germ weight in 100 kg, too, may be of decisive importance in establishing the feeding value of a hybrid.

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### References

- ANASTASIJEVIC, V.—MILEKIC, M.—PEJIC, D. (1978): Uticaj dodavanja lizina, metionina i bakarnog sulfata na povecanje bioloske vrednosti obtoka opaque 2 kukuruz-sojina sacma odnosno. Suncokretova sacma u ishrani odlucene prasadi Stocarstvo, **32**, 109–115.
- ANONYMOUS (1971): Handbuch für den Automatischen Aminosäuren Analysator BC 200. München.
- CARPENTER, K. J. (1960): The estimation of the available lysine in animal-protein foods. Biochem. J., **77**, 604–610.



- CARPENTER, K. J.—BOOTH, U. H. (1973): Damage to lysine in food processing: its measurement and its significance. *Nutr. Abstr. Revs.*, **43**, 423–451.
- CROMWELL, G. L.—PICKETT, R. A.—BEESON, W. M. (1967): Nutritional value of opaque-2 corn for swine. *J. Anim. Sci.*, **26**, 1325–1331.
- EGGUM, B. O.—VILLEGAS, E. M.—VASAL, S. K. (1979): Progress in quality of maize. *J. Sci. Food Agric.*, **30**, 1148–1153.
- HARPSTEAD, D. D. (1971): As a source of protein for men and other nonruminant animals, corn is deficient in the amino acid lysine. This deficiency is now being remedied by the breeding of high-lysine strains. *Scientific American*, **225**, 34–42.
- KLEIN, R. G.—BEESON, W. M.—CLINE, T. R.—MERTZ, E. T. (1971): Opaque-2 and Floury-2 corn studies with growing swine. *J. Anim. Sci.*, **32**, 256–261.
- NOTHEISZ, K.—MARÁZ, L. (1977): Some aspects of zein and lysine determination in maize. *V. Nemzetközi Aminosav Szimpózium*, H.3. Budapest.
- PICK, R. T.—MEADE, R. J. (1971): Amino acid supplementation of opaque-2 corn diets for growing rats. *J. Nutr.*, **101**, 1241–1248.
- RIVERA, P. H.—PEO, E. R.—FLOWERDAY, D.—CRENSHAW, T. D.—MOSER, B. D.—CUNNINGHAM, P. J. (1978): Effect of maturity and drying temperature on nutritional quality and amino acid availability of normal and opaque-2 corn for rats and swine. *J. Anim. Sci.*, **46**, 1024–1036.
- ROSA, J. G.—FORSYTH, D. M.—GLOWER, D. V.—CLINE, T. R. (1977): Normal, opaque-w, waxy, waxy opaque-2, sugary-2 and sugary-2 opaque-2 corn (*Zea mays* L.) endosperm types for rats and pigs. Studies on protein quality. *J. Anim. Sci.*, **44**, 1011–1020.
- SZELÉNYI-GALÁNTAI, M. (1969): Nitrogénforgalmi vizsgálatok a takarmányfehérjék biológiai értékének meghatározására (Nitrogen turnover tests to determine the biological value of feed proteins). *Állattenyésztés*, **18**, 189–191.
- WAHLSTROM, R. C.—MERRILL, R. C.—REINER, L. J.—LIBAL, G. W. (1977): Mutant corns in young pig diets and amino acid supplementation of opaque-2 corn. *J. Anim. Sci.*, **45**, 747–753.

#### COMPARATIVE STUDY ON MAJOR AGRONOMIC CHARACTERISTICS OF MALE FERTILE (NORMAL) AND CYTOPLASMIC MALE STERILE ANALOGUES IN MAIZE (*ZEa MAYS* L.)

An account was given by DUVICK (1965) in a comprehensive study of cytoplasmically male sterile analogues compared to their normal counterparts, from various aspects. In agreement with other authors (JONES—MANGELSDORF 1951, JOSEPHSON—KINCER 1962, JOHNSTON—SNYDER 1962), he found the plant height to decrease under the influence of the T cytoplasm.

The data on the trend of grain yield are contradictory. According to a number of authors the T cytoplasm has no influence on the grain yield (EVERETT 1960, JOSEPHSON—KINCER 1962, JOHNSTON—SNYDER 1962). NOBLE—RUSSEL (1963) pointed out a decrease, while STRINGFIELD (1958) an increase, in grain yield in genotypes with restored T cytoplasm. On the other hand, experiments carried out in various places with different plant numbers unequivocally proved that in the case of a high plant number and unfavourable ecological conditions, the cytoplasmically male sterile hybrids yielded more than their fertile (N-cytoplasm) counterparts (DUVICK 1958, CHINWUBA *et al.* 1961). The authors explained it with the abated competition between ear and tassel. SANFORD *et al.* (1965) arrived at a similar conclusion. In their experiments laid out with high plant numbers, the male sterile genotypes developed a larger number of ears, and the proportion of nitrogen accumulated in the ear, in comparison to the tassel, was also essentially higher in them than that measured in the normal analogues.

As to the other agronomic characteristics (water content of grain on harvesting, breaking of stalk, root lodging, tillering, etc.), steady differences attributable to the T cytoplasm were not found (EVERETT 1960, JOSEPHSON—KINCER 1962, JONES *et al.* 1955).



In our small-plot experiments, we tried to find out what differences in major agronomic characteristics the non-T cytoplasmically male sterile maternal components of some hybrids showed in comparison with their analogues with normal cytoplasm.

In our experiments, maternal parents of some state certified hybrids, prospective varieties producible on a cms basis, obtained from the Cereal Research Institute were used. Thus, in the small-plot field trials, normal and cms analogues of inbred lines (A654, GK71, Szv13, W64A, A632, B37), sister-line crosses (A632  $\times$  A635, W64A  $\times$  WF9) and single crosses (GK71  $\times$  GK72, A654  $\times$  A641, WF9  $\times$  M14, B37  $\times$  A635) were included. (By cms analogue a male sterile specimen produced on a given cms source by backcrossing the recurrent parent at least seven times is understood.) In the case of several genotypes, two different cytoplasmic male sterile analogues of the normal form were also included in the experiment.

Each of the cms analogues examined — except A632cms-C/Rb — showed complete male sterility. Considering the different competitive abilities of the lines and crosses, we set both the inbred lines and their cms analogues (14 genotypes), and the crosses and their cms analogues (14 genotypes) in separate part experiments.

The trials were laid out in 1980 at two ecologically different sites — on the Ságvári grounds of the Institute at Szeged and in its Research Station at Táplánszentkereszt — with four and three replications, respectively, in a random block design. We sowed at optimum time in both places, by means of a sowing gun, 3 kernels per hill, of the same experimental seed lot. The row space was 70 cm, the depth of sowing 6–8 cm, the size of plot 5 m<sup>2</sup>. Stand density was adjusted to 6 plant/m<sup>2</sup> thinning at the stage of 5–6 leaves.

The time of a 50 per cent female flowering was determined by surveying every two days. The yield of each plot was harvested by hand successively, in accordance with the differences in ripening time, but — naturally — the cms analogues always simultaneous with their fertile counterparts. Before the harvest the number of plants broken below the ear as well as the number of ears per plot were recorded. The water content of kernel was determined from ear segments (10 ears/plot) in Szeged and from the total yield of a plot at Táplánszentkereszt, on the basis of loss of weight after drying. The thousand-kernel-weight was determined from 4  $\times$  250 kernels. The data were evaluated by variance analysis.

The soil of the experimental area at Szeged was a highly calcareous meadow chernozem with a humus layer of 60 cm average depth, and solonetz underlying. The pH of the soil was 7.4; it was well supplied with P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O. The annual amount of fertilizer was 400 kg/ha mixed active ingredient (N : P : K = 2 : 1 : 1).

At Táplánszentkereszt the soil of the experimental area was a medium heavy loam, with a 45 cm thick top-soil, 5.8–6.0 pH and good P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O status. The annual supply of fertilizer was 500 kg/ha mixed active ingredient (N : P : K = 1.5 : 1 : 1). Maize had been grown for several years in monoculture at both sites.

At Szeged the weather during the vegetation period was characterized by substantially less than usual sunshine and lower temperature, and by an unevenly distributed though not much less precipitation. In June, for example nearly half of the monthly amount of rain fell in a single day, while in July rainfalls were so scarce that damages by drought were caused. Owing to the low number of sunshine hours and cold weather in May and June, the progress of vegetation was slow, the growth season became 2–3 weeks longer.

All in all, we can say that the weather in 1980 at Szeged was unfavourable compared to the average of many previous years.

At Táplánszentkereszt — as in all parts of the country — the weather during the vegetation period was characterized by less sunshine and lower temperatures; the amount of precipitation, on the other hand, exceeded the historical average. The growth season was considerably longer here too, but, owing to the abundant rainfalls, the yield was more favourable than at Szeged, although the late genotypes were harvested with a very high water content.

Table 1

*Comparative study of some agronomic characteristics of six inbred lines and their cms analogues (Szeged, Táplánszentkereszt, 1980)*

Lines	Grain yield, kg/10 m <sup>2</sup>		Number of days from sowing to 50%♀ flowering		Water content of grain		Breaking of stalk (%)	
	Szeged	Táplánszent- kereszt	Sz	T	Sz	T	Sz	T
A654	3.45	4.07	80.75	94.67	28.3	32.5	0.9	3.4
A654cms-R	5.16**	5.60**	77.75**	93.33	28.7	24.5**	3.1	0.0
A654cms-C	4.55**	5.39**	79.75	94.00	27.3	25.9**	1.8	2.4
GK71	2.99	3.34	73.00	87.67	17.4	21.0	54.6	48.5
GK71cms-C	3.14	3.53	74.00	87.33	18.7	21.4	54.7	56.4*
Szv13	4.78	4.61	86.25	96.00	25.4	26.4	1.7	0.0
Szv13cms-ML	5.60**	5.32*	84.75**	96.00	25.0	21.0**	0.9	1.1
W64A	4.97	3.33	90.75	103.00	28.5	38.7	3.3	5.6
W64Acms-R	5.26	4.54**	90.00	101.67	28.2	40.5	0.9	12.7
W64Acms-ML	7.23**	5.88**	87.50**	99.67**	25.1**	32.9**	5.9	15.8**
A632	5.83	4.68	88.00	103.00	25.8	37.7	2.6	1.1
A632cms-Rb	6.97**	6.26**	88.00	101.00*	24.7	33.6**	0.0	2.3
B37	2.68	2.35	92.75	104.00	32.8	51.8	0.0	1.1
B37cms-C	5.14**	5.22**	91.26**	102.33*	32.4	46.5**	1.1	0.0

\* The difference between the normal form and its male sterile analogue is significant at P5%.

\*\* The difference between the normal form and its male sterile analogue is significant at P1%.

The cms analogues of the six inbred lines exceeded in grain yield the normal counterparts at both experimental sites (Table 1). In 8 cases of comparison the difference was found to be significant 6 times at Szeged and 7 times at Táplánszentkereszt. As regards the number of days from sowing to 50 per cent female flowering, the difference between the two experimental sites was nearly two weeks on an average. At the same time, differences between the sterile and fertile analogues occurred nearly to the same extent at both sites. The male sterile genotypes generally reached the stage of 50 per cent female flowering sooner than, or at the same time as, the normal counterparts. The B37cms-C and W64Acms-ML were earlier than their normal analogues at both sites. The A632cms-Rb proved earlier only at Szeged, while the A654cms-R and Szv13cms-ML at Táplánszentkereszt alone.

The water content of grain on harvesting was substantially higher at Táplánszentkereszt, in accordance with the difference in flowering time (Table 1). Here the water content of grain on harvesting was significantly lower in the A654 male sterile analogues, the Szv13cms-ML, W64Acms-ML and B37cms-C than in the normal analogues, while at Szeged only the W64Acms-ML showed a significant difference. As for the breaking of stalk consistent differences were not found; the extent of stalk breaking was only significant in GK71cms-C and W64Acms-ML at Táplánszentkereszt.



Table 2

*Comparative study on the relative grain yield and number of ear/plant in six inbred lines and their cms analogues (Szeged, Táplánszentkereszt, 1980)*

Lines	Grain yield (%)		Number of ear/plant		Thousand-kernel-weight (g)	
	Sz	T	Sz	T	Sz	T
A654	100	100	0.81	0.86	237	226
A654cms-R	149**	138**	0.97**	0.96	263**	246
A654cms-C	132**	132**	0.89	0.95	241	237
GK71	100	100	0.97	0.97	192	185
GK71cms-C	105	106	1.00	1.02	179	176
Szv13	100	100	0.97	0.97	214	179
Szv13cms-ML	117**	115*	1.01	0.97	220	184
W64A	100	100	0.94	0.91	268	226
W64Acms-R	106	129**	0.93	0.96	272	220
W64Acms-ML	145**	167**	0.93	0.94	285*	219
A632	100	100	1.40	1.53	276	254
A632cms-Rb	120**	134**	1.86**	1.82**	230**	230*
B37	100	100	1.28	1.10	298	250
B37cms-C	192**	222**	1.49**	1.28*	323	287**

\* The difference between the normal form and its male sterile analogue is significant at P5%.

\*\* The difference between the normal form and its male sterile analogue is significant at P1%.

As regards the number of ear/plant, significant differences were observed in favour of the genotypes A632cms-Rb, B37cms-C and A654cms-R. With the latter male sterile form, the difference was reliable only at Szeged (Table 2). In thousand-kernel-weight the A632cms-Rb was significantly inferior to its normal analogue at both sites. On the other hand, the thousand-kernel-weights of A654cms-R and W64Acms-ML were significantly larger at Szeged, while at Táplánszentkereszt the male sterile analogue of B37 proved superior to the normal counterpart in this parameter.

The quantitative indices of some agronomic characteristics of single-crosses, sister-line crosses and their cms analogues are shown in Tables 3 and 4.

Between the sterile and fertile analogues — apart from two exceptions — no reliable differences were found either in stalk breaking or in the water content of grain at the time of harvesting. The exceptions were the higher water content of grain in A654cms-C×A641 (Szeged) and the larger proportion of stalk breaking in GK71cms-C×GK72 (Táplánszentkereszt) (Table 3). Significant difference concerning the grain yield was obtained with two genotypes. At Szeged the male sterile forms of the A632×A635 sister-line cross, while at Táplánszentkereszt the male sterile form of the B37×A635 basic single-cross, yielded more than the normal analogue. As regards the number of days from sowing to the time of 50 per cent female flowering, the male sterile forms proved earlier on an average than the male fertile



Table 3

*Comparative study on some agronomic characteristics in basic single-crosses, sister-line crosses and their cms analogues (Szeged, Táplánszentkereszt, 1980)*

Basic single crosses — Sister-line crosses	Grain yield, kg/10 m <sup>2</sup>		Number of days from sowing to 50%♀ flowering		Water content of grain (%)		Breaking of stalk (%)	
	Sz	T	Sz	T	Sz	T	Sz	T
GK71×GK72	6.34	6.82	71.00	83.33	26.9	32.2	19.8	7.9
GK71cms-C×GK72	6.19	6.57	70.75	82.33	27.8	31.2	14.4	18.2**
A654×A641	9.33	11.08	74.50	89.33	31.6	31.8	1.8	0.0
A654cms-R×A641	8.92	10.73	75.50	89.33	30.7	31.8	0.9	3.3
A654cms-C×A641	9.37	11.58	77.25**	89.67	33.8*	33.4	0.9	3.4
A632×A635	5.75	7.53	88.25	99.67	26.8	36.5	8.0	1.1
A632cms-C×A635	7.34**	8.21	86.50**	98.00*	27.0	34.5	6.9	0.0
A632cms-Rb×A635	7.00*	8.42	87.25	99.67	26.4	36.1	6.2	1.2
W64A×WF9	8.42	10.05	89.00	98.67	29.1	40.3	3.4	0.0
W64Acms-R×WF9	8.08	10.09	87.75*	99.33	28.6	39.1	8.7	0.0
WF9×M14	10.30	14.09	91.50	99.67	29.3	35.0	18.5	2.3
WF9cms-S×M14	10.31	13.79	87.00**	97.67*	29.4	34.1	16.8	5.9
B37×A635	10.82	12.59	85.75	97.33	32.1	33.9	0.0	1.1
B37cms-C×A635	11.81	13.74*	85.00	99.00*	31.6	34.6	2.7	0.0

\* The difference between the normal form and its male sterile analogue is significant at P<sub>5</sub>%.

\*\* The difference between the normal form and its male sterile analogue is significant at P<sub>1</sub>%.

analogues. Reliable difference was shown, however, only by A632cms-C×A635 and WF9cms-S×M14 (at both sites), and W64Acms-R×WF9 (at Szeged). Two male sterile forms — A654cms-C×A641 (at Szeged) and B37cms-C×A635 (at Táplánszentkereszt) —, on the other hand, flowered significantly later.

Remarkable correlation can be observed between the relative grain yield and the number of ear/plant. Increase in the grain yield seems to be measurable first of all in those male sterile hybrids (A632cms-C/Rb×A635, B37cms-C×A635) which, in comparison to the normal analogues, develop a second ear more frequently (Table 4). Significant difference in thousand-kernel-weight was shown in a positive direction by A654cms-R×A641 (at Szeged), while in a negative direction by W64Acms-R×WF9 (at Szeged) and B37cms-C×A635 (at Táplánszentkereszt).

Knowing the data we can thus establish that it was in the extent of stalk breaking, of all agronomic characteristics examined that the lowest difference was found between the normal and male sterile analogues. In 32 cases of comparison (16 sterile-fertile analogous pairs at two sites) it happened only three times that significant differences were obtained. In two of these cases, the difference was caused by the poorer stalk strength of GK71cms-C.

Considerable differences were shown, on the other hand, in the grain yield. The male sterile analogues either exceeded the male fertile forms, or reliably did not yield less than

Table 4

*Comparative study on relative grain yield, number of ear per plant and thousand-kernel-weight in basic single-crosses, sister-line crosses and their cms analogues (Szeged, Táplánszentkereszt, 1980)*

Basic single-crosses, sister-line crosses	Grain yield (%)		Number of ear/plant		Thousand-kernel-weight (g)	
	Sz	T	Sz	T	Sz	T
GK71×GK72	100	100	1.00	1.07	302	317
GK71cms-C×GK72	98	96	1.04	1.04	303	312
A654×A641	100	100	0.97	1.13	326	320
A654cms-R×A641	96	97	1.00	1.11	327	337*
A654cms-C×A641	101	104	0.98	1.14	320	317
A632×A635	100	100	1.01	1.38	263	266
A632cms-C×A635	128**	109	1.30**	1.52	263	272
A632cms-Rb×A635	122*	112	1.32**	1.63**	260	259
W64A×WF9	100	100	0.99	0.99	273	230
W64Acms-R×WF9	96	100	0.93	1.00	260*	238
WF9×M14	100	100	0.95	1.29	277	252
WF9cms-S×M14	100	98	0.90	1.18	270	249
B37×A635	100	100	1.06	1.20	293	283
B37cms-C×A635	109	109*	1.16	1.58**	293	239**

\* The difference between the normal form and its male sterile analogue is significant at P<sub>5%</sub>.

\*\* The difference between the normal form and its male sterile analogue is significant at P<sub>1%</sub>.

those. With the differences considered according to lines and crosses, the above statement is valid with the following specification: while the steady increase in yield is a typical property of the cms line analogues, a yield level similar to that of the normal forms is characteristic of the cms analogues of single-crosses and sister-line crosses. Of the latter genotypes, A632cms-C/Rb×A635 and B37cms-C×A635 are exceptions, as they yielded significantly more than their normal analogues either at one or at the other experimental site. In the genotypes the significant increase in yield can be explained with the larger number of ear/plant.

The question arises, why only in these genotypes do the male sterile forms develop more than one ear. The reason — in our opinion — is that the normal forms of both A632×A635 and B37×A635 also tend to develop several ears per plant. The manifestation of the tendency is supposed to be controlled by the amount of incorporable assimilates. The assimilative productivity, on the other hand, greatly depends on ecological factors. With all this taken into consideration, it is easy to understand that the normal cytoplasm forms of the above crosses developed more frequently a second ear at Táplánszentkereszt — where the ecological conditions are more favourable — than at Szeged. This also confirms our earlier experiences, namely, that the genotypes are really of “prolific” type. In the increase of ear number per plant in their male sterile analogues, on the other hand, a change in the proportions of incorporable assimilates rather than an increase in their absolute quantity is sup-

posed to be the regulatory factor. The higher relative proportion of assimilates available for the grain yield is in this case a logical consequence of the abated competition between tassel and ear primordia (GROGAN *et al.* 1965).

The steady increase in yield observed in the cms line analogues is in all probability due to the decreased competition as well. The relative excess of incorporable assimilates resulting from this is here of special importance, since, in the maize lines, the photosynthesis is not so intensive as in the hybrids (BÁLINT 1977). The great difference in yield between the normal and C-cytoplasm analogues of B37 can, however, be traced back not only to this, but also to the late flowering and deficient seed-set of the normal form.

As regards the number of days from sowing to the time of 50 per cent female flowering, the male sterile forms proved in most cases earlier than the male fertile analogues, with two exceptions showing reliable differences: A654cms-C  $\times$  A641 (at Szeged) and B37cms-C  $\times$  A635 (at Táplánszentkereszt) which exceeded their normal counterparts in the water content of grain, too.

Steadily great differences in the water content of grain on harvesting were found only at Táplánszentkereszt, between the normal and cms analogues of lines. The water content of grain in the male sterile line analogues was far below that in the male fertile counterparts. On the basis of the data of the Szeged trial, we assume that, in the case of harvesting with a lower water content of grain, the differences decrease. Reliable differences in the number of ear/plant occurred only in those cms analogues whose normal, male fertile forms also tend to develop more than one ear. As to the thousand-kernel-weight, the differences do not show any permanent tendency.

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### References

- BÁLINT, A.: Gazdasági növényeink produkciós genetikája (Production genetics of crops in Hungary). Akadémiai Kiadó, Budapest, 1977.
- CHINWUBA, D. M.—GROGAN, C. O.—ZUBER, M. S. (1961): The action of detasseling, sterility and spacing on yields of corn hybrids. *Crop Sci.*, **1**, 279–280.
- DUVICK, D. N. (1958): Yields and other agronomic characteristics of cytoplasmically pollen sterile corn hybrids compared to their normal counterparts. *Agron. J.*, **50**, 121–125.
- DUVICK, D. N. (1965): Cytoplasmic pollen sterility in corn. *Advan. Genet.*, **13**, 1–56.
- EVERETT, H. L. (1960): Effect of cytoplasm and Rf gene in maize. *Agron. J.*, **52**, 215–216.
- GROGAN, C. O.—PATRICIA SARVELLA—SANFORD, J. O.—JORDAN, H. V. (1965): Influence of cytoplasmic male sterility on dry matter accumulation in maize (*Zea mays* L.). *Crop Sci.*, **5**, 365–367.
- JOHNSTON, G. S.—SNYDER, R. J. (1962): The utilization of cytoplasmic male sterility for sweet corn hybrid seed production. *Amer. Soc. Hort. Sci.*, **81**, 415–420.
- JONES, D. F.—MANGELSDORF, P. C. (1951): The production of hybrid seed corn seed without detasseling. *Connecticut Agr. Exp. Sta. Bull.*, 550.
- JONES, D. F.—STINSON, H. T.—MUNSON, A. P.—RENSHAW, C. C. (1955): Field and sweet corn report for 1954. Mount Carmel, Conn. Report of progress-GI Dept. of Genetics Connecticut Agr. Exp. Sta., New Haven, Conn. Mimeograph. Feb.
- JOSEPHSON, L. M.—KINCER, H. C. (1962): Effects of male sterile cytoplasm on yield and other agronomic characteristics of corn inbreds and hybrids. *Crop Sci.*, **2**, 41–43.
- NOBLE, S. W.—RUSSEL, W. A. (1963): Effects of male sterile cytoplasm and pollen fertility restorer genes on performance of hybrid corn. *Crop Sci.*, **3**, 92–96.
- SANFORD, J. O.—GROGAN, C. O.—JORDAN, H. V.—SARVELLA, P. A. (1965): Influence of male sterility on nitrogen utilization in corn, *Zea mays* L. *Agron. J.*, **57**, 580–583.
- STRINGFIELD, G. H. (1958): Fertility restoration and yield in maize. *Agron. J.*, **50**, 215–218.



# CHARACTERISTICS OF DIGITALIS LANATA EHRH. ROSETTES OF DIFFERENT GROWTH TYPES

The *Digitalis lanata* Ehrh. is an important industrial drug plant. Beyond its quantitative and qualitative properties (KUTIÁK 1977, MASTENBROEK 1978) its suitability for mechanical harvesting is today also an important aspect in the work of breeding; namely, the leaves of the variety to be produced should be erect.

In the course of our work, we tried to find out what was the percentage of plants with erect leaves in the *D. lanata* population, what on the one hand were their histological characteristics, and whether on the other the leaf- and lanatoside C production of plants of this habit was favourable.

In a strain produced by selection from the variety *Digitalis lanata* Ehrh. cv. Oxfordi (NYÁRÁDI 1973), we studied 301 plants chosen at random at rosette stage, at the end of the vegetative phase of development.

Field examinations. a) The growth habit of the rosette and the number of leaves were recorded for each plant.

Histological examinations. b) Fully developed leaves were collected from plants with different growth habits to study the tissue structure of leaf. The fresh leaves were rinsed with water and fixed in a mixture of formalin-alcohol-acetic acid (FAA) in the usual way (JOHANSEN 1940). From the basal-, middle- and apical part of the leaf, cross-sections were taken. These were frozen in a mixture of gelatine and water (1 : 1). For pre-freezing, carbone dioxide gas and Anderson—Green—Wood et Co.'s Quick freeze chamber were used. Sections of 13  $\mu$ m were prepared in a MIRKÖZ cryostat and stained with a 0.20 per cent solution of toluidine blue; an equal ratio mixture of 2% potassium iodide- and 2% ferro(III)-cyanide solution was used as chelating agent (ROMHÁNYI 1968) and a 1 : 1 mixture of glycerine and water to cover the sections. The preparations were framed with nail polish. The microscope examinations were carried out with a Reichert Zetopan research microscope. Ten leaves of each type were measured as shown in Fig. 1.

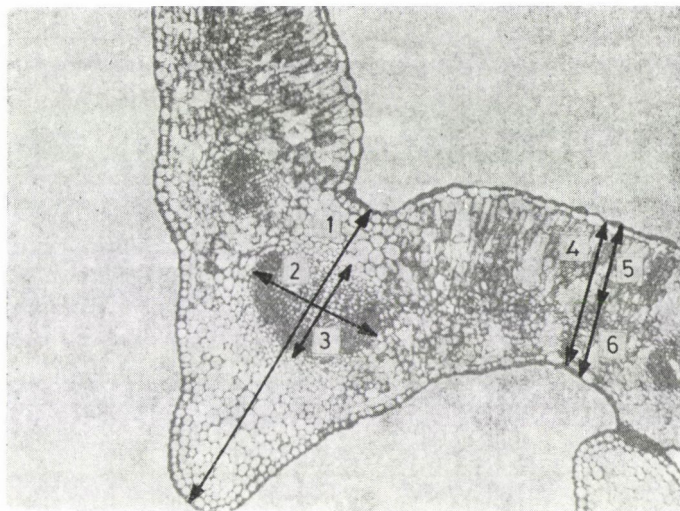


Fig. 1. Measuring data in the cross-section of *Digitalis lanata* Ehrh. leaf ( $\times 25.2$ ). 1. Thickness of leaf along the main rib, 2. "width" of main rib, 3. "height" of main rib, 4. thickness of leaf blade, 5. thickness of palisade parenchyma, 6. thickness of spongy parenchyma



Fig. 2. *Digitalis lanata* Ehrh. rosette of erect habit



Fig. 3. *Digitalis lanata* Ehrh. rosette of normal habit

Chemical analyses. c) For the determination of the lanatoside C content, samples of fifteen leaves each were collected and examined with the method described by SZILÁGYI *et al.* (1977). The data were evaluated with biometric methods (SVÁB 1973).

In spite of the contradictory results, favourable experiences were gained with some important crops — rice, wheat, barley, soya, maize — in the past decades (HAYASHI—Ito 1962, PENDLETON 1968); varieties with erect leaves were found to grow more vigorously and produce larger yields than the normal types. PIOVARIC—VIDOVIC (1971) pointed out for maize that, by negative selection, higher yields were obtained from progenies of erect leaf types, and lower yields from those of horizontal leaf types.

*D. lanata* is a biennial plant with a rosette in the first year. The rosette is low, with reclinate leaves; the lower leaves lie on the ground surface and become severely infected and





Fig. 4. *Digitalis lanata* Ehrh. rosette of procumbent habit

mostly decay by the time of harvesting. The leaf is of bifacial structure. In cross-section the epidermis cells are isodiametrical, the stomata slightly protrude. The palisade parenchyma consists of 3–4 and the spongy parenchyma of 6–7 cell rows. The mesophyll is not differentiated into spongy- and palisade parenchyma, either at the leaf base or along the main rib. Below the main rib the walls of parenchyma cells, close to the abaxial epidermis, are slightly colenchymatic. The vascular bundles are collateral.

According to our investigations in the Oxfordi strain population, three growth types could be distinguished. Of the population 13 per cent (39 plants) had erect leaves (*erectum*), most favourable from the point of view of mechanical harvesting and stand density (Fig. 2); 87 per cent (260 plants) were of normal type (with bent or reclinate leaves) (Fig. 3), and 0.006 per cent (2 plants) procumbent (Fig. 4). The histological characteristics of procumbent plants are not given here, since they occurred in a negligible proportion in the population.

The examination results of the tissue structures of rosettes of normal and erect habit are summed up in Table 1. In the different habit plants the number of cell rows in the palisade and spongy parenchyma was the same, and agreed with the values of the general characterization. According to our examinations, the cells in the leaves of rosettes of erect habit were larger, as proved by the fact that in the erect rosettes the full thickness of leaf along the main rib as well as leaf thickness reduced by the height of the main rib were larger at a significance level of  $P = 5\%$  at the leaf base,  $P = 1\%$  in the middle of leaf and  $P = 0.1\%$  at the leaf apex, compared with the normal plants.

The ratio of width to height of the main rib at the leaf base and in the middle of leaf is not only significantly higher in the erect than in the normal type, but their values are also identical. In the case of normal plants, on the other hand, it is in the middle of the leaf and at the leaf apex that this ratio is the same. The ratio of palisade to spongy parenchyma is, at the level of significance, different in the erect plants; at the leaf apex, significantly lower than in the normal type. As for the thickness of the leaf blade, the mean values of plants with erect leaves are higher, though the difference is not mathematically demonstrable.

Considering the leaf output of the population parts studied, we found that plants with a normal habit had an average leaf number of 110.3 and those with erect leaves, 105.4. The latter value is lower, but the difference is not significant. In the erect type part of the



**Table 1**  
*Histological characteristics of leaves of different habit rosettes*

Characteristics	Habit	Leaf base		Middle of leaf		Leaf apex	
		$\bar{x}$	SD	$\bar{x}$	SD	$\bar{x}$	SD
Full thickness of leaf along the main rib*	Erect	174.3	24.73	101.7	14.81	48.4	5.74
	Normal	144.7	24.00	77.6	16.70	37.6	5.58
	SD5%	22.9		14.8		5.3	
	SD1%	—		20.3		7.3	
	SD0.1%	—		—		9.9	
Leaf thickness reduced by the height of the main rib*	Erect	121.1	19.62	64.4	9.41	30.9	3.18
	Normal	98.0	20.09	47.7	14.06	22.7	4.55
	SD5%	18.7		11.2		3.7	
	SD1%	—		15.4		5.1	
	SD0.1%	—		—		6.9	
Ratio of width to height of the main rib	Erect	2.1	0.62	2.1	0.60	1.2	0.23
	Normal	1.4	0.30	1.1	0.10	1.1	0.10
	SD5%	0.5		0.4		—	
	SD1%	—		0.6		—	
	SD0.1%	—		0.9		—	
Thickness of leaf blade*	Erect	47.0	9.74	38.4	3.53	33.1	5.11
	Normal	42.0	4.61	37.3	4.85	30.1	3.63
Ratio of palisade to spongy parenchyma	Erect	—		1.15	0.36	1.56	0.34
	Normal	—		1.48	0.39	1.94	0.37
	SD5%	—		—		0.33	

\* Figures multiplied by 12.3 give the values in  $\mu\text{m}$ .

population the mean value of leaf number in 18 plants exceeded in tendency that of the normal type ( $\bar{x} = 115.8$ ).

As regards the lanatoside C content, the result is similar. The mean value of lanatoside C content in plants with erect leaves is lower by some 20 per cent than in the normal type, but in seven plants it exceeds the mean value of the normal type, and the number of leaves in these is also the best in the whole population ( $\bar{x} = 147$ ). With the leaf number and the lanatoside C content jointly taken into consideration, 18 per cent of the erect type part of the population, 2.3 per cent of the full population — a total of 7 plants, apart from those of a growth habit suitable for mechanical harvesting, have large leaf yields and favourable lanatoside C contents. Since we have found specimens showing correlations, there is a possibility of combining several favourable characteristics. We are going to continue studying the part of population isolated on the ground of our examination results, and extend our investigations by including other populations.

According to our investigations, the population of *D. lanata* Ehrh. cv. Oxfordi studied by us could be divided into two morphologically highly different groups: 87 per cent of the plants were of normal, 13 per cent of erect growth habit. The tissue structure of plants with erect leaves was also significantly different from that of the normal type.

Of the erect-leaf part of population, suitable for mechanical harvesting 18 per cent exceeded the normal type part of the population by 30 per cent in leaf number, and by 20 per cent as regards lanatoside C content.

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### References

- HAYASHI, K.—ITO, H. (1962): Studies on the form of plant in rice varieties with particular reference to the efficiency in utilizing sunlight. *Proc. Crop Sci. Soc. Japan*, **30**, 329–333.
- JOHANSEN, D. A. (1940): *Plant microtechnique*. McGraw-Hill Book Company, Inc. New York—London.
- KUTIAK, A. F. (1977): Versuchsanbau von *Digitalis lanata* (nach einem Vortrag auf dem Internationalen Arzneipflanzen Kollegium Juli 1976 in Graz Steiermark, Österreich) *Hgk. Mitteilungen*, **20**, 135–139.
- MASTENBROEK, C. (1978): Aus der Arbeit "Kruidentuin Doornspijk", Niederlande, *Hgk. Mitteilungen*, **21**, 45–47.
- NYÁRÁDI-SZABADY, J. (1973): *Digitalis lanata* Ehrh. törzsek glikozidtartalmának és összetételének változása nemesítés hatására (Changes in the glycoside content and composition of *Digitalis lanata* Ehrh. strains under the influence of breeding). *Herba Hung.*, **12**, 65–71.
- PENDLETON, I. W. (1968): Light relationship and corn plant geometry. In: *Proc. of the 23th Annual Corn and Sorghum Research Conf., ASTA, Chicago* 91–96.
- PROVARIČ, A.—VIDIVIČ, I. (1973): Leaf angle in corn (*Zea mays* L.) breeding. In: *Proc. of the 7th Meeting of Maize and Sorghum Section Eucarpia — Zagreb* 11.
- ROMHÁNYI, GY. (1968): A citomembránok ultrastruktúrájáról (Ultrastructure of cytomembranes). *MTA Biol. Oszt. Közl.*, **11**, 127–149.
- SVÁB, J. (1973): Biometriai módszerek a mezőgazdasági kutatásban (Biometric methods in agricultural research). *Mezőgazdasági Kiadó, Budapest*.
- SZILÁGYI, I.—ZÁMBÓ, I.—TÖRÖK, J. (1977): Eine serienanalytische Methode der Lanatosid C und deren exakte Beweise. *Planta Med.*, **32**, 60–67.

### CHARACTER ASSOCIATION AND PATH ANALYSIS IN SORGHUM

*Sorghum* is an important food and feed crop cultivated both in the monsoon (Kharif, June—July planting) and the winter (Rabi, September—October planting) seasons. The winter sorghum crop depends on residual soil moisture and the yield is mainly a function of monsoon rains. Association studies supply reliable information on the nature, extent and directions of selection. A knowledge of genetic correlation becomes of paramount importance when the breeder is dealing with quantitative characters in a crop like sorghum which is cultivated in two seasons. The studies of genetic correlations and path analysis are limited and mostly reported by evaluating the material in the monsoon season (SINGH—BAGHEL 1977, DABHOLKAR *et al.* 1970). The present studies of genetic correlations and path analyses were therefore conducted during both monsoon and winter seasons.

The material for the present investigation consisted of eighty hybrids developed by crossing eight male sterile lines (2219A, CK60A, 1036A, 2077A, 36A, 1202A, 1258A and 3660A) with ten restorer parents (IS-84, 3924, CS-3541, PD-3-1-11, 168, 285, 1235, 1324, 370 and 358). These 80 hybrids and 18 parents were evaluated in a randomized block design with three replications during the monsoon and winter seasons of 1976-77 at Parbhani. Each genotype was sown in a three metre long single row with 15 cm and 45 cm spacing within and between the rows, respectively. Recommended cultivation practices were adopted to grow a good crop. The data were recorded on ten competitive plants in each replication. The observations were recorded on nine characters, viz. plant height (cm), days to 50% flowering, panicle length (cm), panicle weight (g), number of whorls/panicle, number of primary branches/panicle, number of secondary branches/panicle, 10 $\bar{a}$ -grain weight (g) and grain yield/plant (g).

Genotypic correlations were calculated and path analyses were carried out for each season after DEWEY—LU (1959).

Estimates of genotypic correlation coefficients between various traits are given in Table 1. The correlations were significant for 21 and 15 of 28 character pairs in the monsoon and winter seasons, respectively. Plant height is positively and significantly correlated with all characters except panicle length in the monsoon season. However, plant height under winter conditions exhibited a significant positive association only with panicle length, panicle weight, number of whorls/panicle and grain weight. Days to flowering gave a significant positive association with five characters in the monsoon season. Panicle length and grain size gave significant positive and negative associations, respectively, with days to flowering in the winter season. Number of whorls (panicle, number of primaries) (panicle and number of secondaries) panicle revealed significant positive correlations with most of the traits in both

Table 1  
*Genotypic correlations between different pairs of characters under monsoon and winter conditions in sorghum*

Characters		Days to flowering	Panicle length (cm)	Panicle weight (g)	Number of whorls per panicle	Number of primaries per panicle	Number of secondaries per panicle	100 grain weight (g)
Plant height (cm)	K	0.375**	-0.027	0.398**	0.487**	0.277**	0.363**	0.419**
	R	-0.091	0.348**	0.266**	0.380**	0.087	0.187	0.341**
Days to flowering	K		-0.147	0.234**	0.140	0.371**	0.493**	0.243**
	R		0.320**	-0.052	0.097	0.100	0.217	-0.408**
Panicle length (cm)	K			-0.114	0.256*	0.041	0.111	-0.180
	R			0.173	0.512**	0.181	0.369**	-0.109
Panicle weight (g)	K				0.493**	0.341**	0.423**	0.589**
	R				0.540**	0.267**	0.350**	-0.034
Number of whorls per panicle	K					0.287**	0.389**	0.327**
	R					0.385**	0.801**	-0.201*
Number of primaries per panicle	K						0.994**	0.305**
	R						0.863**	-0.111
Number of secondaries per panicle	K							0.311**
	R							-0.204

K = *Kharif* or Monsoon, R = *Rabi* or Winter

\*, \*\* Significant at 5% and 1% respectively



Table 2

Path coefficient analysis showing the direct and indirect effects of eight characters on grain yield under monsoon and winter conditions in sorghum

Characters		Effects via								Genotypic correlation with grain yield
		Plant height (cm)	Days to flowering	Panicle length (cm)	Panicle weight (g)	Number of whorls per panicle	Number of primaries per panicle	Number of secondaries per panicle	100 grain weight (g)	
Plant height (cm)	K	0.064	−0.021	−0.001	0.374	−0.031	−0.071	0.108	0.018	0.440**
	R	−0.006	0.010	0.031	0.271	−0.106	−0.032	0.097	0.016	0.281**
Days to flowering	K	0.024	−0.057	−0.004	0.220	−0.009	−0.096	0.148	0.010	0.236*
	R	0.001	−0.115	0.027	−0.053	−0.027	−0.037	0.112	−0.020	−0.112
Panicle length (cm)	K	−0.002	0.008	0.032	−0.107	−0.016	−0.010	0.033	−0.008	−0.070
	R	−0.002	−0.037	0.090	0.176	−0.144	−0.068	0.191	−0.005	0.201*
Panicle weight (g)	K	0.025	−0.013	−0.003	0.941	−0.032	−0.088	0.127	0.025	0.982**
	R	−0.001	0.006	0.015	1.019	−0.152	−0.100	0.180	−0.001	0.966**
Number of whorls per panicle	K	0.031	−0.008	0.008	0.464	−0.065	−0.074	0.117	0.014	0.487**
	R	−0.002	−0.011	0.046	0.550	−0.281	−0.145	0.413	−0.009	0.561**
Number of primaries per panicle	K	0.018	−0.021	0.001	0.321	−0.018	−0.259	0.298	0.013	0.353**
	R	−0.001	−0.011	0.016	0.272	−0.108	−0.375	0.445	−0.005	0.233*
Number of secondaries per panicle	K	0.023	−0.028	0.004	0.398	−0.025	−0.257	0.300	0.013	0.428**
	R	−0.001	−0.025	0.033	0.356	−0.225	−0.323	0.516	−0.010	0.321**
100-grain weight (g)	K	0.027	−0.014	−0.005	0.544	−0.021	−0.079	0.093	0.043	0.598**
	R	−0.002	0.047	−0.010	−0.034	0.057	0.041	−0.105	0.049	0.043

K and R = *Kharif* (Monsoon) and *Rabi* (Winter) season respectively

Residual effect: 0.180 (K), 0.252 (R); \* Significant at 5%; \*\* Significant at 1%

seasons. SINGH—BAGHEL (1977) also recorded a positive association between the number of primary and secondary branches.

Grain size gave a significant positive correlation with most of the traits in the monsoon season. The significant negative association of grain size with days to flowering in the winter season may be due to limiting moisture conditions. Grain yield/plant (Table 2) gave a significant positive correlation with most of the characters in the monsoon season. SINGH—BAGHEL (1977) also reported a positive association of yield with the four panicle characters studied. In the present study, however, in the winter season, grain yield exhibited a non-significant correlation with days to flowering and grain weight. These observations indicate that it is essential to develop drought-escaping (early maturing) or tolerating genotypes for winter season cultivation.

Yield is influenced by many factors. Selection based on simple correlations without taking into consideration the interactions between the component characters may be misleading in some situation. A perusal of direct and indirect effects (Table 2) in both the seasons revealed that with respect to panicle weight and number of secondaries per panicle the contribution through direct effects was positive and high in comparison to indirect effects, indicating that these traits should be given more importance when making the selection. Indirectly these two traits also affected yield favourably, indicating no mutual cancelling effect between these characters. DABHOLKAR *et al.* (1970) and SINGH—BAGHEL (1977) also reported high direct effects of the number of grains per panicle and the number of secondaries per panicle on grain yield in the monsoon season.

In the case of panicle length, though the direct effect was positive in the monsoon season, it was nullified by negative indirect effects, thus making the correlation negative. In the winter season, however, the positive direct and indirect effects resulted in a positive correlation of panicle length with grain yield. Grain weight exhibited positive direct effects in both seasons. However, negative indirect effects during the winter season resulted in a weak correlation. These results indicated that correlation and indirect effects in respect of panicle length and 100-grain weight are influenced by the season.

Four characters, viz. plant height, days to flowering, number of whorls per panicle and number of primaries per panicle, gave positive correlations with yield, but the results of path analysis were not encouraging, thereby indicating their insignificant contribution to the yield.

As judged from direct and indirect path effects and correlations over two seasons the characters panicle weight and number of secondaries per panicle showed a stable association with yield, thus indicating the importance of these characters as selection criteria for increasing yield potential in breeding programmes.

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### References

- DABHOLKAR, A. R.—TELANG, S. W.—PATEL, K. C. (1970): Path analysis of yield components in hybrid sorghum. *Indian J. Genet.*, **30**, 625–629.  
 DEWEY, R. D.—LU, K. H. (1959): A correlation and path-coefficient analysis of components of crested wheat grass seed production. *Agron. J.*, **51**, 515–518.  
 SINGH, R. P.—BAGHEL, S. S. (1977): Yield components and their implications to selection in sorghum. *Indian J. Genet.*, **37**, 62–67.

ESTIMATES OF GENETIC VARIABILITY AND INTERRELATIONSHIP  
OF YIELD COMPONENTS IN SAFFLOWER  
(*CARTHAMUS TINCTORIUS* L.)

Safflower is one of the major oilseed crops extensively grown in India. The varieties so far released for cultivation were evolved by the selection system of breeding and are poor yielders (average yield 700–800 kg/ha). This crop, therefore, needs improvement and genetic upgrading. A knowledge of genetic variability is essential before initiating the breeding programme. In safflower this type of information is very limited. ASHRI *et al.* (1974), ARGIKAR *et al.* (1957) and ABEL (1969) studied the association between the yield components. ASHRI *et al.* (1974) studied a collection from India, China and Egypt and concluded that the degree of association between the yield and the yield components was not consistent. This situation, therefore, emphasises the need for further investigation on this aspect.

In the present investigation an attempt has been made to estimate the extent of variability for various plant characters and their association with each other.

Twenty-eight genotypes of safflower were grown in a randomized block design with three replications at three locations during the rabi season of 1977–78. Each plot consisted of three rows each 3 m in length. The rows were spaced at 45 cm and the seeds were dibbled at 15 cm within a row. Five plants in the central row were selected randomly for recording observations on days to maturity, plant height (cm), branches per plant, capitula per plant, seeds per capitulum, 1000-seed mass (g) and seed yield per plant (g).

The genotypic and phenotypic coefficients of variability were estimated according to BURTON (1952). Heritability in the broad sense was worked out as per HANSON *et al.* (1956). The correlation coefficients for the possible combinations of seven characters were calculated.

The results of analysis of variance for seven characters at three locations are presented in Table 1. The results revealed that the genotypes differ significantly at all the locations for all the characters. This clearly indicated the presence of sufficient variability among the genotypes.

**Table 1**

*Analysis of variance for seven characters in safflower under three environments*

Source of variation	Location	Mean sum of squares						
		Days to maturity	Height (cm)	Branches/plant	Capitula/plant	Seeds/capitula	1000-seed mass (g)	Seed yield/plant (g)
Replications	I	2	2.00	39.14	8.11	717.14	13.12	1.02
	II	2	6.15	212.20	12.58	39.96	4.88	4.56
	III	2	10.45	135.59	0.59	345.95	48.67	1.85
Treatments	I	27	44.58**	188.25**	5.46	304.06**	110.36**	528.12**
	II	27	37.57**	222.52**	7.22**	177.76**	137.42**	516.76**
	III	27	40.82**	341.25**	4.14**	152.05*	283.95**	491.33**
Error	I	54	0.77	29.15	3.33	95.40	6.29	2.86
	II	54	2.62	27.42	3.21	63.66	10.80	3.16
	III	54	1.55	16.78	1.62	84.11	12.46	4.06

Note: I = Parbhani, II = Latur and III = Badnapur

\* Significant at 5% level, \*\* Significant at 1% level



Table 2

*Estimates of genetic parameters for seven characters at three locations in safflower*

Genetic parameters	Loca- tion	Characters						
		Days to maturity	Height (cm)	Branches/plant	Capitula/plant	Seeds/capitula	1000-seed mass (g)	Seed yield/ plant (g)
Mean	I	138.79	73.01	11.94	54.96	21.52	55.79	33.01
	II	133.82	81.16	10.78	36.86	26.69	51.39	29.99
	III	138.67	82.22	10.18	40.82	26.55	53.98	36.57
Range	I	124.00 to	53.46 to	9.26 to	40.26 to	13.20 to	29.66 to	13.86 to
		142.66	87.53	14.33	81.93	44.26	72.66	53.20
	II	125.00 to	69.53 to	8.13 to	25.93 to	20.66 to	28.33 to	13.60 to
		139.66	106.46	14.06	57.06	57.66	70.00	45.76
	III	122.00 to	53.26 to	7.60 to	20.00 to	19.80 to	33.66 to	18.40 to
		142.00	106.46	12.20	53.33	71.00	71.00	45.03
Genotypic coefficient of variation (GCV)	I	2.80	10.00	7.10	15.20	27.40	23.70	31.20
	II	2.60	9.90	10.70	16.70	24.30	25.50	25.00
	III	2.60	12.60	9.00	11.70	35.80	23.60	20.20
Phenotypic coefficient of variation (PCV)	I	2.80	12.40	16.80	23.40	29.70	23.90	33.20
	II	2.80	11.80	19.80	27.40	27.30	25.70	34.90
	III	2.80	13.60	15.40	25.30	38.20	23.90	23.60
Heritability broad sense (%)	I	95.00	64.53	17.56	42.16	84.70	98.40	88.20
	II	81.60	70.34	29.36	37.40	79.60	98.20	51.30
	III	89.40	86.57	34.02	21.21	87.90	97.60	72.90
Genetic advance (E)	I	2.35	6.41	12.41	7.64	44.18	17.76	31.71
	II	2.10	6.13	24.96	10.94	32.10	19.95	20.81
	III	2.22	7.99	27.01	5.38	42.15	18.21	19.94

Note: I = Parbhani, II = Latur and III = Badnapur

Estimates for variability parameters for seven characters are shown in Table 2. The mean performance for yield contributing characters like branches per plant, capitula per plant and 1000-seed mass was higher at Parbhani. The Parbhani location therefore appears to be favourable as compared to the other two locations. A wide range of variability was observed for all the characters at all the locations except for 1000-seed mass. The values for the phenotypic coefficient of variability were larger in magnitude at all the locations for all the characters. Three characters, namely seed per capitulum, seed yield per plant and 1000-seed mass gave high values for the genotypic coefficient of variability (25.50 to 35.80). Similarly high values for the genotypic coefficient of variability were reported by THOMBRE—JOSH (1977) for yield per plant and capitulum size. However, low GCV values were recorded for plant height (9.90 to 12.60), branches per plant (7.10 to 10.70) and capitula per plant (11.70 to 15.20). Plant height, seeds per capitulum and 1000-seed mass gave consistent GCV and PCV values across locations. These three characters therefore appear to be resistant to environmental fluctuations. Similar findings were reported by MAKNE *et al.* (1979).

COMSTOCK *et al.* (1958) stated that the relative amount of the heritable portion of variation can be assessed through the heritability percentage. The heritability estimates were high for 1000-seed mass (97.60 to 98.40), days to maturity (81.60 to 95.00), seeds per capitulum

Table 3

*Genotypic correlation coefficients between seven yield components in safflower at three locations*

Characters	Location	Height (cm)	Branches/plant	Capitula/plant	Seeds/capitula	1000-seed mass (g)	Days to maturity	Seed yield/plant (g)
Height (cm)	I		-0.4217*	-0.3876*	0.4807**	0.1187	0.5761**	0.0334
	II		-0.3620	-0.6038**	0.2761	-0.3827*	0.3625	-0.3393
	III		0.2459	-0.3114	0.7546**	0.4677*	0.5663**	0.3775*
Branches/plant	I			0.8748**	-0.5216**	0.3336	-0.0282	0.2020
	II			0.8485**	-0.4271*	0.3151	0.3272	0.2028
	III			0.6277**	-0.3342	0.2336	0.6081**	0.2368
Capitula/plant	I				-0.2631	0.4888**	0.1206	0.3616
	II				-0.3553	0.5691**	-0.0448	0.5654**
	III				-0.6060**	0.4289*	0.3050	0.5326**
Seeds/capitulum	I					0.6518**	-0.0048	0.6613**
	II					0.4557*	0.2576	0.4475*
	III					0.5301**	0.2698	0.4251*
1000-seed mass (g)	I						-0.1212	0.9512**
	II						-0.0388	0.9664**
	III						0.3594	0.9231**
Days to maturity	I							-0.2549
	II							-0.0863
	III							0.3862*

Note: I = Parbhani, II = Latur and III = Badnapur

\* Significant at 5% level, \*\* Significant at 1% level

(79.60 to 87.90), seed yield per plant (51.30 to 88.20) and plant height (64.53 to 86.57). The present findings are in agreement with THOMBRE—JOSHI (1977) for all characters except plant height and with MAKNE *et al.* (1979) for all characters except seed yield per plant. The heritability estimates for all characters except seed yield were consistent over different locations. This indicated the importance of individual plant selection for these characters. Genetic advance was highest (32.10 to 44.18) for seed per capitulum at all the locations, indicating the possibility of maximum genetic progress. MATHER *et al.* (1976) reported a high expected genetic advance for seed yield and seeds per capitulum and medium to low values for other characters.

High heritability estimates coupled with high genetic advance is an indication of the predominance of additive gene effects (PANSE 1957). In the present study characters such as seeds per capitulum, 1000-seed mass and seed yield per plant can thus be said to be governed by additive gene effects. Individual plant selections for these characters should therefore be effective and fruitful in breeding programmes.

The yield per plant was found to be positively and significantly correlated with 1000-seed mass, seeds per capitulum and capitula per plant at all the locations (Table 3). This indicated that yield is directly related with these components. Similar findings were reported by ABEL (1976). ASHRI *et al.* (1974) and MATHER *et al.* (1976) reported a close association between head diameter and yield in safflower. Plant height exhibited a weak association with most of the characters except seeds per capitula. Branches per plant was found to be significantly correlated with capitula per plant, whereas it was negatively and significantly correlated with seeds per capitulum. The negative association of capitula per plant and seeds per capitula at all the locations suggests the use of inter se mating in advanced generations for the desired combinations, as both are important yield contributing characters. Significant and positive correlations were observed between capitula per plant and 1000-seed mass at all the locations. Seeds per capitulum and 1000-seed mass were positively and significantly correlated at all the locations. The association of branches per plant with capitula per plant, and 1000-seed mass with yield were found to be not only stronger but also consistent at all the location.

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### References

- ABEL, G. H. (1969): An analysis of yield components in safflower. Third. Proc. Safflower Res. Conf., University of California, Davis. 18–22.
- ABEL, G. H. (1976): Relationships and uses of yield components in safflower breeding. *Agron. J.*, **68**, 442–447.
- ARGIKAR, G. P.—MORBAD, I. R.—THAMBI, V. V. (1957): The range of variation and correlation of some quantitative characters in *Carthamus tinctorius* L. *Indian Oilseed J.*, **1**, 228–234.
- ASHRI, A.—ZIMMER, D. E.—URIE, A. L.—CAHANER, A.—MARANI, A. (1974): Evaluation of germplasm collection of safflower (*Carthamus tinctorius* L.). IV. Yield and yield components and their relationship. *Crop Sci.*, **14**, 799–803.
- BURTON, G. W. (1952): Quantitative inheritance in grasses. Proc. 6th Inst. Grass Cong., **1**, 277–283.
- COMSTOCK, R. E.—KELLEHER, T.—MORROW, E. B. (1958): Genetic variation in an asexual species, the garden strawberry. *Genetics*, **43**, 634–646.
- HANSON, G. H.—ROBINSON, H. F.—COMSTOCK, R. E. (1956): Biometrical studies on yield in segregating population of Koreah Lespedeza. *Agron. K.*, **48**, 268–272.



- MAKNE, V. G.—PATIL, V. D.—CHOUDHARI, V. P. (1979): Genetic variability and character association in safflower. *Indian J. Agric. Sci.*, **49**, 766–768.
- MATHER, J. R.—TIKKA, G. B.—SHARMA, R. K.—SINGH, S. P.—DASHORA, S. L. (1976): Genetic variability and Path-coefficient analysis of yield components in safflower. *Indian J. Hered.*, **8**, 1–9.
- PANSE, V. G. (1957): Genetics of quantitative characters in relation to plant breeding. *Indian J. Genet.*, **17**, 318–328.
- THOMBRE, M. V.—JOSHI, B. P. (1977): A biometrical approach to selection problems in safflower (*Carthamus tinctorius* L.) varieties. *J. Maharashtra Agril. Univ.*, **2**, 1–3.

## LEAF THINNING EFFECT AND CORRELATION STUDY BETWEEN CURD WEIGHT AND CERTAIN OTHER CHARACTERS IN CAULIFLOWER

In recent years fodder has become very expensive. This has made farmers in Egypt think of using any green plant or leaves as nutriment for their animals. From the vegetable crops accordingly used, the cauliflower gives the largest quantity of leaves. Therefore, research was carried out to discover the effect of leaf thinning and the characters responsible for a good yield.

Cauliflower seeds of the Orgival Selected cultivar were used in two successive seasons (1974–75 and 1975–76) in the Vegetable Research Centre of the Faculty of Agriculture, Cairo University at Giza in A. R. Egypt.

The treatments used were: a) Thinning the mature leaves every week till two weeks before harvesting. b) Thinning 6 mature leaves one month before harvesting and 4 more leaves during the next week. c) Thinning 10 mature leaves at one time, three weeks before harvesting. d) Leaving the plants without thinning as a control.

A complete randomized block design was used in four replicates.

In the 1974–75 season, the seeds were sown in a seedbed on August 5th 1974 and transplanted to the field on September 17th 1974; the harvest was on January 28th 1975. In the second season (1975–76) the seeds were sown in the seedbed on August 1st 1975, transplanted on September 4th 1975 and harvested on January 22nd 1976.

Data were recorded on the same day of harvest in both years for the following characters:

### I. Effect of leaf thinning on plant characteristics

1. Stem length and diameter in cm. The stem diameter was measured in the cross-section in the upper part of the stem after removing the curd.

2. Leaf number and weight. The weights were measured in kilogrammes.

3. Curd characteristics

a) Curd weight in kg.

b) Length and diameter in cm.

c) Shape index =  $\frac{\text{length}}{\text{diameter}}$

4. Ratio of foliage weight to curd weight (F : C ratio) =  $\frac{\text{foliage weight}}{\text{curd weight}}$

### II. Correlation

Data on a population of 63 plants were used in the 1975–76 season to reveal the association between characters. Simple and partial correlation of the first, second and third (compound) order were used. The degree of association between quantitative characters was determined by calculating the coefficient of correlation.

The calculations were carried out using the methods suggested by SNEDECOR—COCHRAN (1967).

### *I. Effect of leaf thinning*

The combined data for the effect of leaf thinning on the following characters are shown in Table 1.

#### 1. Stem characteristics:

- a) Length: The short stem was characteristic of the continuous weekly leaf thinning, while the other treatments were similar in length.
- b) Diameter: The largest diameter was found in normal plants and the smallest after weekly leaf thinning; the other treatments were intermediate.

#### 2. Leaf characteristics:

- a) Number: The normal plants had the most leaves, followed by those thinned once or twice, and then by those thinned weekly.
- b) Weight: The trend was the same as for the number of leaves.

#### 3. Curd characteristics:

- a) Weight: The heaviest curds were found in normal plants (without thinning), the smallest weight were found with continuous weekly thinning, and the other two treatments were intermediate.
- b) Diameter and length: Normal plants and those thinned once or twice were nearly the same for diameter and length of curd, while the curds of plants thinned every week were much smaller.
- c) Shape index: There were no significant differences between the different treatments used and all the curds were flattened to about the same extent.

#### 4. Ratio of foliage weight to curd weight (F : C ratio):

No significant differences were observed between the various treatments.

From the data given above, leaf thinning had a deleterious effect on plant characteristics, resulting in a decrease in the length and diameter of the stem, the number and weight of the leaves and the weight of the curd, especially in the weekly thinning treatment. Thus, thinning the leaves of cauliflower plants is not recommended.

There is no record in the available literature on thinning the leaves of cauliflower.

**Table 1**

*Effect of leaves thinning on the plant characteristics*  
(Combined analysis of years 1974–75 and 1975–76)

Characters  Treatments	Stem		Leaves		Curd			Foliage to curd weight	
	length	diam- eter	No.	weight, kg	weight, kg	diam- eter	length	shape index	(F : C) ratio
	cm					cm			
Without leaves thinning (control)	26.5	4.2	27.8	4.127	2.416	28.7	19.4	0.68	1.95 : 1
Leaves thinning once	27.2	4.0	22.6	3.418	2.143	28.3	19.3	0.65	1.83 : 1
Leaves thinning twice	26.4	3.9	22.7	3.252	2.072	27.4	18.3	0.69	1.80 : 1
Continuous weekly leaves thinning	18.7	3.5	13.6	0.795	0.557	17.3	11.9	0.70	1.72 : 1
L.S.D. (0.05)	1.9	0.3	1.8	0.293	0.174	1.3	1.8	N.S.	N.S.

### III. Correlations

Correlations were calculated to throw light on the degree of relationship or association between any two of the studied characters, especially between curd weight and other characters which influence the curd weight. Thus, all these sorts of correlations were taken from the data of control plants (without leaf thinning) in the season 1975-76.

#### A) Single correlation:

Thirty-six correlations were computed between the following pairs of characters, as summarized in Table 2.

I. Positive, highly significant correlation coefficients ( $> +0.5$ ) were found between the following characters:

1. Curd weight and leaf weight.
4. Curd weight and curd diameter.
5. Curd weight and curd length.
8. Curd weight and stem diameter.
10. Leaf weight and leaf number.
11. Leaf weight and curd diameter.
15. Leaf weight and stem diameter.
26. Leaf number and stem diameter.
27. Curd diameter and curd length.
30. Curd diameter and stem diameter.

II. Significant or highly significant positive correlation coefficients ( $< +0.5$ ) were found between:

3. Curd weight and leaf number.
12. Leaf weight and curd length.
14. Leaf weight and stem length.
22. Leaf number and curd diameter.
23. Leaf number and curd length.
24. Leaf number and shape index of curd.
25. Leaf number and stem length.
33. Curd length and stem diameter.

III. On the other hand, a highly significant negative correlation coefficient ( $> -0.5$ ) was obtained between:

28. Curd diameter and shape index of curd.

IV. Significant or highly significant negative correlation coefficients ( $< -0.5$ ) were obtained between:

2. Curd weight and F : C ratio.
6. Curd weight and shape index of curd.
13. Leaf weight and shape index of curd.
16. F : C ratio and curd diameter.
17. F : C ratio and curd length.
35. Shape index and stem diameter.

V. There were no significant correlations between the other characters under investigation.

#### B) Partial and compound correlations:

The simple correlations show that there are four characters which influence the curd weight. Thus, 70 partial correlations of the first, second and third order (compound) in ten groups (Table 3) were studied between the following five characters to determine which characters had a direct effect on curd weight:



1. Curd weight.
2. Leaf weight.
3. F : C ratio.
4. Leaf number.
5. Stem diameter.

1. A positive, highly significant correlation existed between curd weight and leaf weight when the effect of F : C ratio, leaf number or stem diameter was eliminated (A-1, 2

Table 2

*Values of single correlation coefficient (r) obtained for nine characters in the Orgival Selected cultivar of cauliflower*

Characters correlated		r
1	Curd weight and leaf weight	+0.629**
2	Curd weight and F : C ratio	-0.497**
3	Curd weight and leaf number	+0.470**
4	Curd weight and curd diameter	+0.795**
5	Curd weight and curd length	+0.683**
6	Curd weight and shape index	-0.307*
7	Curd weight and stem length	+0.113 <sup>n.s.</sup>
8	Curd weight and stem diameter	+0.663**
9	Leaf weight and F : C ratio	+0.104 <sup>n.s.</sup>
10	Leaf weight and leaf number	+0.535**
11	Leaf weight and curd diameter	+0.616**
12	Leaf weight and curd length	+0.477**
13	Leaf weight and shape index	-0.306*
14	Leaf weight and stem length	+0.332**
15	Leaf weight and stem diameter	+0.647**
16	F : C ratio and leaf number	-0.026 <sup>n.s.</sup>
17	F : C ratio and curd diameter	-0.404**
18	F : C ratio and curd length	-0.428**
19	F : C ratio and shape index	+0.057 <sup>n.s.</sup>
20	F : C ratio and stem length	+0.243 <sup>n.s.</sup>
21	F : C ratio and stem diameter	-0.189 <sup>n.s.</sup>
22	Leaf number and curd diameter	+0.283*
23	Leaf number and curd length	+0.259*
24	Leaf number and shape index	+0.440**
25	Leaf number and stem length	+0.304*
26	Leaf number and stem diameter	+0.611**
27	Curd diameter and curd length	+0.703**
28	Curd diameter and shape index	-0.554**
29	Curd diameter and stem length	+0.056 <sup>n.s.</sup>
30	Curd diameter and stem diameter	+0.544**
31	Curd length and shape index	+0.188 <sup>n.s.</sup>
32	Curd length and stem length	-0.001 <sup>n.s.</sup>
33	Curd length and stem diameter	+0.445**
34	Shape index and stem length	-0.078 <sup>n.s.</sup>
35	Shape index and stem diameter	-0.259*
36	Stem length and stem diameter	+0.151 <sup>n.s.</sup>

and 3) or when the effect of both F : C ratio and leaf number (A-4), F : C ratio and stem diameter (A-5), or number of leaves and stem diameter (A-6) was constant, or when the effect of F : C ratio, leaf number and stem diameter was eliminated (A-7).

Table 3

*Partial correlation of the first, second and third order (compound)  
in Orgival Selected population*

(1. Curd weight. 2. Leaf weight. 3. F : C ratio, 4. No. of leaves. 5. Stem diameter)

Groups	Correlations	r values in the population	Groups	Correlations	r values in the population
A 1	12.3	+0.7887**	F 1	24.1	+0.3489**
2	12.4	+0.5062**	2	24.3	+0.5408**
3	12.5	+0.3504**	3	24.5	+0.2314n.s.
4	12.34	+0.7045**	4	24.13	+0.2396n.s.
5	12.35	+0.6118**	5	24.15	+0.2073n.s.
6	12.45	+0.3362**	6	24.35	+0.2076n.s.
7	12.345	+0.5954**	7	24.135	+0.1132n.s.
B 1	13.2	-0.7274**	G 1	25.1	+0.3952**
2	13.4	-0.5494**	2	25.3	+0.6826**
3	13.5	-0.5056**	3	25.4	+0.4786**
4	13.24	-0.7261**	4	25.13	+0.3405**
5	13.25	-0.6849**	5	25.14	+0.2839*
6	13.45	-0.5248**	6	25.34	+0.5269**
7	13.245	-0.6875**	7	25.134	+0.2723*
C 1	14.2	+0.2033n.s.	H 1	34.1	+0.2711*
2	14.3	+0.5269**	2	34.2	-0.0971n.s.
3	14.5	+0.1095n.s.	3	34.5	+0.1151n.s.
4	14.23	+0.1942n.s.	4	34.12	+0.0756n.s.
5	14.25	+0.0312n.s.	5	34.15	+0.1988n.s.
6	14.35	+0.1957n.s.	6	34.25	+0.0487n.s.
7	14.235	+0.0888n.s.	7	34.125	+0.0965n.s.
D 1	15.2	+0.4318**	I 1	35.1	+0.2163n.s.
2	15.3	+0.6679**	2	35.2	-0.3379**
3	15.4	+0.5378**	3	35.4	-0.2187n.s.
4	15.23	+0.2882*	4	35.12	-0.0385n.s.
5	15.24	+0.3903**	5	35.14	+0.1090n.s.
6	15.34	+0.5121**	6	35.24	-0.3284**
7	15.234	+0.2338n.s.	7	35.124	-0.0713n.s.
E 1	23.1	+0.6176**	J 1	45.1	+0.4531**
2	23.4	+0.1396n.s.	2	45.2	+0.4112**
3	23.5	+0.3022*	3	45.3	+0.6175**
4	23.14	-0.1922n.s.	4	45.12	+0.3662**
5	23.15	+0.5932**	5	45.13	+0.4199**
6	23.45	+0.2852*	6	45.23	+0.4039**
7	23.145	-0.2341n.s.	7	45.123	+0.3704**

2. A highly significant negative correlation was observed between curd weight and F : C ratio with the elimination of the effect of leaf weight, leaf number or stem diameter (B-1, 2 and 3), and also when the effect of both leaf weight and leaf number (B-4), leaf weight and stem diameter (B-5) or leaf number and stem diameter (B-6) was constant, and finally, when leaf weight, leaf number and stem diameter were constant (B-7).

3. A non-significant correlation was found between curd weight and leaf number in all cases (C-1, 3, 4, 5, 6 and 7) except when the effect of the F : C ratio was eliminated, in which case there was a highly significant positive correlation (C-2).

4. A highly significant or significant positive correlation was evident between curd weight and stem diameter in all cases (D-1, 2, 3, 4, 5 and 6), except that a non-significant correlation was recorded when the effect of leaf weight, F : C ratio and leaf number was eliminated (D-7).

5. A significant or highly significant positive correlation was detected between leaf weight and F : C ratio when the effect of curd weight or stem diameter was eliminated (E-1 and 3) and when both curd weight and stem diameter or leaf number and stem diameter (E-5 and 6) were constant. The remaining cases were not significant (E-2, 4 and 7).

6. A non-significant correlation was noticed between leaf weight and leaf number in all cases except when the effect of curd weight or F : C ratio was constant (F-1 and 2), when the correlation was highly significant.

7. A highly significant or significant positive correlation existed between leaf weight and stem diameter when curd weight, F : C ratio or leaf number was constant (G-1, 2 and 3) as well as when the effects of both curd weight and F : C ratio (G-4), curd weight and leaf number (G-5), or F : C ratio and leaf number (G-6) were eliminated. The same result was found in the last case, when the effects of curd weight, F : C ratio and leaf number were eliminated collectively (G-7).

8. A non-significant correlation was obtained between F : C ratio and leaf number in all cases (H-2, 3, 4, 5, 6 and 7), except that this relation was significant when curd weight was eliminated (H-1).

9. A non-significant correlation was recorded between F : C ratio and stem diameter in most cases (I-1, 3, 4, 5 and 7). However, a highly significant negative correlation existed between these traits when the effect of leaf weight (I-2) or of both leaf weight and leaf number (I-6) was constant.

10. A highly significant positive correlation was observed between leaf number and stem diameter when one, two or three of the remaining characters was unaffected (J-1, 2, 3, 4, 5, 6 and 7).

From the above correlation results, there was a highly significant positive correlation coefficient between curd weight and leaf weight when the effect of the other characters was eliminated, but the number of leaves did not affect the curd weight in the compound correlation. This was because the leaves are the site where photosynthesis is accomplished, the products of which are transmissible to the buds to form the curd. As a result of this fact, if the leaves are heavier in weight, the curd also becomes heavier. Thus, the weight of the leaves is more important than the number. This result was also proved by the fact that the compound correlation between leaf weight and leaf number in this investigation was insignificant.

Naturally, there was also a highly significant negative correlation between curd weight and F : C ratio when the other characters were constant. In other words, if the leaf weight was heavier than normal, the result would be a smaller curd. Therefore, to obtain a heavy curd, the weight of the leaves must increase to a certain limit, after which the curd decreases in weight.



In spite of the clear effect of stem diameter on leaf weight and number in all the correlations, it had no effect on the curd weight in partial correlations of the third order.

No record was found in the available literature on correlation between the above mentioned characters in cauliflower, Egypt

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### References

SNEDECOR, G. W.—COCHRAN, W. S. (1967): Statistical methods. The Iowa State University Press, Ames, Iowa, U.S.A. 593.

### INFLUENCE OF GROWTH REGULATING CHEMICALS ON YIELD AND QUALITY OF TWO PLANT TYPES COTTON

The main object of using growth regulating chemicals is to attain balanced vegetative growth and fruiting, resulting in higher yields. Slight changes in the climatic factors, soil and cultural practices leading to varying behaviour of the plants resulted in unstable productivity, as evidenced by a variation of 107 to 155 kg/ha average yield over 5 years (ANONYMOUS 1975). These growth stimulating and growth retarding chemicals have been known to enhance the effects of agrotechniques and can increase the efficiency of the costly inputs, leading to high, stable production.

The investigations were carried out on the two plant types of cotton (*Gossypium hirsutum* L. and *Gossypium arboreum* L.) in a pot culture during 1973–74 and in the field during 1974–75.

#### Pot culture experiment

This experiment was conducted in pots and included two periods of seed soaking, i.e. for 6 hr and 9 hr before sowing. There were 13 treatments: Dry seed (C), Water soaking (CW), soaking the seeds in  $M^{-5}$ ,  $M^{-4}$  and  $M^{-3}$  concentrations of succinic acid ( $S_1S_2S_3$ ), citric acid ( $C_1C_2C_3$ ) and malic acid ( $M_1M_2M_3$ ) and soaking the seed in 500 ppm and 1000 ppm cycocel ( $CS_1CS_2$ ). Two separate experiments were conducted on hirsutum and arboreum cottons. Pots measuring  $23 \times 30$  cm were filled with a mixture of soil and farmyard manure at the rate of 3 : 1. The sowing was done with the help of an adjustable depth peg on 29th May 1973. The plants were raised with optimum water and nutrient conditions under the natural photoperiod prevailing in this region. Observations on the effect of these acids were made on the number of bolls, and the yield per plant.

#### Field experiment

The experiment was conducted at the Research Farm, Haryana Agricultural University, Hissar, during the Kharif season of 1974–75 with 16 treatments of seed soaking for 6 hr and spraying with organic acids and cycocel on two cultivars of cotton, H 14 and G 27, from *Gossypium hirsutum* L. and *Gossypium arboreum* L., respectively. The experiment was laid out in a split plot design in three replications with main plots allotted to organic acids and cycocel and sub-plots to the cultivars. The treatments included seed soaking with water

(CW), varying concentrations ( $M^{-5}$ ,  $M^{-4}$ ,  $M^{-3}$ ) of the organic acids, succinic acid ( $S_1S_2S_3$ ), citric acid ( $C_1C_2C_3$ ) and malic acid ( $M_1M_2M_3$ ), and cycocel (250, 500 and 1000 ppm) ( $CS_1CS_2CS_3$ ), including 3 additional treatments of 100 ppm cycocel spray 45, 60 and 75 days ( $Cy_1Cy_2Cy_3$ ) after sowing. The data obtained on number of bolls per plant, boll weight, seed cotton yield and quality traits was subjected to analysis of variance.

### *Number of bolls/plant*

After seed soaking in organic acids for the two periods both the varieties retained a greater number of bolls per plant than dry seed, water-soaked control or seed soaked in cycocel. There was not much difference between soaking the seeds in water and cycocel. In general, soaking the seeds for 6 hr gave a higher number of bolls than seed soaking for 9 hr. Lower concentrations of all the organic acids proved superior to higher concentrations (Table 1). The total number of bolls decreased with an increase in the concentration of cycocel. In the field experiment, the boll number per plant in H 14 due to various treatments showed less variation than in G 27 (Table 2). Though all the organic acids increased the boll number over the control, the highest increase was obtained with malic acid. Seed soaking in cycocel and cycocel spray significantly reduced the number of bolls per plant in the mean of both varieties when compared with malic acid (Table 2). However, the differences between other treatments were non-significant. As regards the effect of various concentrations, in H 14 the middle concentration of malic and citric acid and in G 27 the lowest concentration of malic acid resulted in the highest number of bolls per plant. The increased number of bolls per plant due to acid treatments might be conjectured from a combination of factors, such as size of

**Table 1**  
*Effect of different treatments on total number of bolls/plant and yield/plant (g)*

Treatments	Total number of bolls/plant				Yield/plant (g)			
	H 14		G 27		H 14		G 27	
	6 hr	9 hr	6 hr	9 hr	6 hr	9 hr	6 hr	9 hr
C	9.0	9.0	12.0	12.0	25.15	25.15	18.30	18.30
CW	11.0	9.5	14.0	12.5	30.80	27.00	21.09	18.87
$S_1$	13.0	12.0	21.0	15.5	37.42	34.58	32.87	24.48
$S_2$	12.5	11.5	23.0	16.0	35.44	32.40	35.80	25.05
$S_3$	11.0	10.0	16.0	15.0	31.30	28.81	24.66	23.14
$C_1$	15.5	12.0	19.5	19.0	44.20	34.20	30.25	28.99
$C_2$	13.5	13.0	17.5	17.0	39.42	37.93	27.45	26.29
$C_3$	11.5	11.0	17.5	16.0	32.81	33.95	27.31	24.81
$M_1$	14.5	13.0	22.0	17.0	41.75	37.41	33.45	25.66
$M_2$	12.5	12.5	20.5	17.0	35.61	35.62	31.72	26.43
$M_3$	12.0	12.5	20.5	18.0	34.08	35.62	31.26	27.54
$CS_1$	11.5	10.5	16.5	14.0	34.28	31.17	26.48	22.47
$CS_2$	9.5	9.5	13.5	11.5	28.54	28.54	21.52	18.34
C.V.	9.73	16.9	20.0	10.1	9.98	16.67	18.34	
S.E.M.	0.830	1.345	2.54	2.132	2.45	4.06	3.62	4.67
L.S.D.5%	2.53	N.S.	7.75	6.50	7.45	12.37	11.01	N.S.

plant, to accomodate a greater number of bolls per unit area and better utilization of the nutrients taken up. Seed soaking in higher concentrations of cycocel or spraying at early stages had a more adverse effect on the number of bolls which matured. This was brought about by reduced growth, low dry matter and the disturbance of the biological sink. The development of the stem is checked and less positions are available for the production of flowers in the small number of sympodia present. Another factor contributing to the lower number of bolls produced might be that the period of boll production was curtailed in plants treated with cycocel. Vegetative growth that precedes flowering might be the factor which played a role in the lower number of bolls with cycocel spray treatment (EASTON 1931).

### *Boll weight*

The boll weight of H 14 was significantly higher than that of G 27, which may be ascribed to the different genetic behaviour of the two. The boll size was not influenced by soaking the seeds in various organic acid solutions (Table 2). However, seed soaking in cycocel and its spray gave a marginal increase in boll weight over the control. This means that boll development enjoyed priority over vegetative growth. The ability of the leaves to manufacture and supply carbohydrates to the developing bolls was promoted by cycocel treatments. These findings are in conformity with the results of SINGH—SINGH (1970).

**Table 2**

*Total number of bolls/plant and boll weight (g) as influenced by certain plant acids and cycocel (1974–75)*

Treatments	Boll weight			Number of bolls/plant		
	H 14	G 27	Mean	H 14	G 27	Mean
CW	2.77	1.51	2.14	26.00	28.20	27.10
S <sub>1</sub>	2.77	1.56	2.16	26.20	28.87	27.28
S <sub>2</sub>	2.79	1.48	2.13	27.35	30.40	28.87
S <sub>3</sub>	2.82	1.47	2.14	26.60	30.50	28.55
C <sub>1</sub>	2.82	1.56	2.19	27.10	31.00	29.05
C <sub>2</sub>	2.82	1.48	2.15	28.20	30.45	29.32
C <sub>3</sub>	2.90	1.53	2.21	26.80	28.88	27.84
M <sub>1</sub>	2.84	1.60	2.21	27.20	31.80	29.50
M <sub>2</sub>	2.93	1.58	2.25	28.00	29.40	28.70
M <sub>3</sub>	2.80	1.58	2.19	27.50	32.00	29.75
CS <sub>1</sub>	2.83	1.61	2.22	26.20	29.00	27.85
CS <sub>2</sub>	2.91	1.59	2.25	26.00	27.20	26.60
CS <sub>3</sub>	2.88	1.61	2.25	25.40	27.00	26.20
Cy <sub>1</sub>	3.03	1.60	2.31	24.40	25.00	24.70
Cy <sub>2</sub>	3.18	1.60	2.39	26.30	25.50	25.77
Cy <sub>3</sub>	2.85	1.58	2.21	26.50	26.70	26.60
	Var.	Treat- ments	Var. × treatments	Var.	Treatments	Var. × treatments
S.E.M.	0.027	0.074	0.110	0.39	1.37	1.59
L.S.D.5%	0.55	N.S.	N.S.	0.796	N.S.	N.S.



**Table 3**  
*Yield of seed cotton (q/ha) as influenced  
 by certain organic acids and cycocel*

Treatments	H 14	G 27	Mean
CW	18.62	19.44	19.03
S <sub>1</sub>	18.45	19.66	19.05
S <sub>2</sub>	20.55	22.10	21.32
S <sub>3</sub>	18.86	23.85	21.35
C <sub>1</sub>	19.30	21.06	20.18
C <sub>2</sub>	22.69	22.20	22.44
C <sub>3</sub>	18.84	20.24	19.54
M <sub>1</sub>	19.98	25.04	22.51
M <sub>2</sub>	22.59	21.22	21.90
M <sub>3</sub>	18.30	26.66	22.48
CS <sub>1</sub>	19.79	22.42	21.10
CS <sub>2</sub>	19.09	18.28	18.68
CS <sub>3</sub>	18.28	17.00	17.64
Cy <sub>1</sub>	16.66	15.49	16.07
Cy <sub>2</sub>	19.54	15.66	17.60
Cy <sub>3</sub>	20.06	18.78	19.42
	Variety	M × S	Treatments
S.E.M.	0.484	1.938	1.441
L.S.D. 5%	N.S.	N.S.	N.S.

#### *Seed cotton yields*

In the pot experiment the productivity per plant in H 14 was higher than in G 27. Various organic acids had a favourable effect on the yield of seed cotton per plant. Malic acid showed superiority over citric and succinic acids (Table 1). In H 14, cycocel did not affect the yield adversely, whereas in G 27 the yield with cycocel decreased. The mean values show that yield per plant (Table 3) increased with a corresponding increase in the concentration of malic acid and vice versa with succinic acid, whereas no definite trend could be observed with citric acid. In cycocel spray treatments, the decrease in yield was greater when spraying was done early in the season. In H 14, the middle concentrations of citric acid, malic acid and succinic acid registered an increase in yield over the water-soaked control of about 4.03, 3.97 and 2.93 q/ha respectively. Spraying at 60 and 75 days after sowing gave a higher yield than in the control, whereas cycocel spraying at early stages gave a lower yield than the control. Seed soaking in a lower concentration of cycocel slightly improved the yield over the control, but higher concentrations gave yields below that of the control (Table 3). In G 27, malic acid gave a higher yield than the control. The yield of seed cotton was reduced by soaking in higher concentrations of cycocel and spraying at early stages. The greater yield of seed cotton after 6 hr soaking than after 9 hr soaking (Table 1) might be attributed to the rapid growth of the plant under the 6 hr period of soaking. The yielding ability of G 27, the *arboresum* cotton, was found to be less than that of H 14, a *hirsutum* type, having a greater boll size that more

Table 4

*Effect of certain organic acids and cycocel on quality characters*

Treatments	Ginning %			Halo length (cm)			Seed index (%)		
	H 14	G 27	Mean	H 14	G 27	Mean	H 14	G 27	Mean
CW	32.8	37.6	35.2	23.33	13.40	18.37	7.24	4.95	6.09
S <sub>1</sub>	33.3	37.5	35.4	23.46	13.93	18.70	7.21	5.62	6.41
S <sub>2</sub>	33.2	37.6	35.4	23.66	13.40	18.53	7.44	5.21	6.32
S <sub>3</sub>	33.3	37.4	35.3	23.80	13.66	18.73	7.39	5.45	6.42
C <sub>1</sub>	33.0	37.7	35.3	23.40	13.83	18.61	7.31	5.45	6.38
C <sub>2</sub>	33.5	37.7	35.6	23.53	14.00	18.77	7.79	5.01	6.40
C <sub>3</sub>	33.4	37.3	35.3	23.66	13.40	18.53	7.93	5.34	6.63
M <sub>1</sub>	33.1	37.5	35.3	23.50	14.06	18.80	7.37	5.52	6.44
M <sub>2</sub>	33.3	37.6	35.4	23.93	13.70	18.81	7.97	5.27	6.62
M <sub>3</sub>	33.5	37.8	35.7	23.53	13.66	18.60	7.60	5.82	6.71
CS <sub>1</sub>	33.7	37.7	35.7	23.93	14.27	19.10	7.63	5.51	6.57
CS <sub>2</sub>	33.5	38.0	35.7	24.23	14.20	19.21	7.46	5.77	6.61
CS <sub>3</sub>	33.8	37.8	35.8	24.20	14.33	19.27	6.93	5.41	6.17
Cy <sub>1</sub>	33.7	37.7	35.7	23.93	14.40	19.07	6.91	5.42	6.16
Cy <sub>2</sub>	33.6	37.8	35.7	24.33	14.30	19.33	7.43	5.37	6.40
Cy <sub>3</sub>	33.9	38.0	35.9	24.00	14.20	19.10	8.02	5.47	6.74
	Var.	M × S	Treatments	Var.	M × S	Treatments	Var.	M × S	Treatments
S.E.M.	0.106	0.54	0.308	0.166	0.655	0.428	0.066	0.264	0.153
L.S.D.5%	0.216	N.S.	N.S.	0.339	N.S.	N.S.	0.134	N.S.	N.S.

than compensated for the contribution made by higher boll number in G 27. The higher yield per plant with citric and malic acid may be ascribed to their favourable effect on growth. Increased yields caused by organic acids have also been recorded before (YAKUBOV—KADYOV 1963, SHALIMOV 1965, BAZANOVA 1967). The positive association between yield and dry matter of different parts of both the cultivars (MALIK *et al.* 1978) indicated an increase in either dry matter or height, or simultaneously in both these characters, as evidenced by the fact that these organic acids brought a corresponding increase in the yield. The yield reduction due to seed soaking in higher concentrations of cycocel or spraying in the early stages might be because growth is checked at an early stage, which is an active period for developing the active sites available for developing bolls. The results are in close agreement with the work of KHAN *et al.* (1968) and ZUR *et al.* (1970, 1972).

#### *Quality of cotton*

Non-significant variations due to different concentrations of various acids and cycocel have been observed in the present investigations. Both seed soaking in different concentrations of cycocel or spraying at different stages had an increasing trend on ginning percentage and halo length in both the cultivars. This might be due to the continuous supply of photosynthates, hormones and nutrients for fibre development (Table 4). The favourable effect of cycocel on fibre quality is in close agreement with the findings of BHATT—NATHAN (1968) and BHATT—RAMANUJAM (1970).

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### References

- ANONYMOUS (1975): Bulletin on cotton. Directorate of Cotton Development, Ministry of Agriculture, Bombay, 9, 5-6.
- BAZANOVA, T. B. (1967): Effect of succinic and naphthenic acids on the five-fibered cotton plant after various doses of nitrogen-phosphorus fertilizers. *Izv. Akad. Nauk Turkm. SSR, Ser. Biol. Nauk.*, 4, 24-29.
- BHATT, J. G.—NATHAN, A. R. S. (1968): Effect of cycocel, phosphon and brino on growth and yield of cotton (*G. hirsutum* L.). *Indian J. Pl. Physiol.*, 11, 225-230.
- BHATT, J. G.—RAMANUJAM, T. (1970): Effect of cycocel on yield, chlorophyll content and fibre properties of lint of MCU cotton. *Indian J. Pl. Physiol.*, 13, 113-115.
- EASTON, F. M. (1931): Root development as related to character of growth and fruitfulness of cotton plant. *Jour. Agri. Res.*, 21, 875-883.
- KHAN, M.—SIRAJ-UJ-DIN—SULTAN, S. C.—WAHEED, B. E. (1968): Use of cycocel growth retardant in cotton. *Pak. J. Agric. Res.*, 6, 52-59.
- MALIK, R. K.—SINGH, C.—SINGH, H.—HOODA, R. S. (1978): Correlation analysis of growth yield and yield characters of two plant types of cotton. *Indian Journal Pl. Physiology*, 21, 90-92.
- SHALIMOV, A. G. (1965): Experiments in the agrolaboratory of the State Farm, Khlokkovodstvo, 15, 54.
- SINGH, H. G.—SINGH, B. (1970): Preliminary studies on the effect of cycocel on cotton (*G. arboreum* L.). *Indian J. Agric. Sci.*, 40, 562-565.
- YAKUBOV, A. M.—KADYVOV, SH. K. (1963): Trace nutrients increase the cotton yield and quality. *Khlopkovodstvo*, 8, 48-50.
- ZUR, M.—MARANI, A.—CARMELI, R. (1970): Effect of CMH (N-dimethyl-N) (B-chloroethyl hydrazonium chloride) as compared with that of CCC (2-chloroethyl trimethyl ammonium chloride) on height, earliness and yield of cotton. *Israel J. Agric. Res.*, 20, 133-134.
- ZUR, M.—MARANI, A.—KARADAVID, B. (1972): Effect of growth retardants CCC and CMH on cotton. *Cott. Grow Rev.*, 49, 250-257.

### AGROTECHNICAL POSSIBILITIES OF CONTROLLING THE CHLOROPID FLIES

The basis of protection from the chloropid flies has for a long time been represented by the properly applied agrotechnics. The procedures elaborated have mostly been concentrated on the autumn damages caused by the gout fly (*Chlorops pumilionis* Bjerk.), the frit fly (*Oscinella frit* L.) and the late wheat shoot fly (*Phorbia securis* Tiensohn). The possibilities of controlling them through cultural practices were already discussed at the turn of the century by MEZEI (1899) and JABLONOWSKI (1901), and the question continued to engage the researchers' attention (RAPAICS 1914, KADOCSA 1923, 1943, KERÉKES 1924, GYÖRFFY 1927, JABLONOWSKI 1927, 1928, ANONYMOUS 1928). On the basis of their investigations, the agrotechnical control can be summarized as follows: 1. Attraction sowing must be completed by the beginning of September and worked on in time. 2. Sowing must not begin before 4 October. 3. On determining the germ number to be sown, damages by flies should be taken into consideration.

The ethological knowledge of the species in question has confirmed the above statements. However, according to recent investigations, the spring flies are becoming more and more important (SÁRINGER 1950, JERMY 1953a, b, JERMY—SZELÉNYI 1958). In spring the



wheat bulb fly (*Delia coarctata* Fall.), the early wheat shoot fly (*Phorbia haberlandti* Schiner.), the spring generation of the late wheat shoot fly (*Phorbia securis* Tiensuu) and the spring generation of the grass and cereal fly (*Opomisa florum* Fabr.) cause damages, as pointed out by DARVAS *et al.* (1981) and SZEŐKE (1981). The most dangerous spring fly pest for winter wheat is the wheat bulb fly. In Hungary, KÜKEDI (1975), KUPAI (1971, 1973) and SZEŐKE (1981) equally called attention to its menace. In Europe it has so far been observed in the following countries: Germany (FRANK 1900), Denmark (ROSTRUP 1911), England (GEMMIL 1923), Sweden (MIEGROET 1950), Austria (FABER 1968), France (RECAMIER 1964), Romania (PERJU—PÉTERFY 1970), Holland (ANONYMOUS 1971), Belgium (ANONYMOUS 1971), Hungary (KUPAI 1972), Czechoslovakia (POKORNY 1972), Poland (MENDE 1974), Norway (MASKELL—DAVIS 1974), European Soviet Union (MASKELL—DAVIS 1974), Switzerland (MEYER 1976). The authors are of the same opinion insofar as the best protection against spring fly pests is the strengthening of cereals before the winter sets in (JERMY 1953a, b). Because of differences in ethology among the species, spring and autumn damages are judged differently. Besides the chemical control, an agrotechnical method of protection from spring flies had to be found as well. Our investigations covered the effects of forecrop, sowing time and implements on the number of population.

a) *Forecrop*. The role of crop rotation in plant protection was pointed out in Hungary by JABLONOWSKI (1928). A number of foreign authors (KLEINE 1918, GEMMIL 1923, GOUGH 1953, 1957, BOLLOW 1960, BUHL 1963, LUTZE—MENDE 1972, KUPAI 1978) think it important to take the ethology of chloropid flies — first of all of the wheat bulb fly — into consideration when determining the succession of crops. According to their investigations after pea, potato, sugar-beet and clovers, the wheat bulb fly is likely to cause considerable damages because of the favourable conditions of egg laying. To prove the validity of the statement under Hungarian conditions, we studied the trend of infestation by wheat bulb fly larvae as a function of the forecrop. In 1978 a trial was set up in five winter fields of the "Vörösmarty" Co-operative Farm, Kápolnásnyék, Lake Velence, sown after potato, sunflower, maize, pea and winter wheat, respectively, as forecrops. Damages done by wheat bulb flies were recorded on two occasions every week. The results are shown in Table 1. Accordingly, no essential differences were observed depending on the forecrop. Egg laying by the female wheat bulb flies obviously forms the basis of infestation in the following year. Therefore, the soil of the forecrop has to be considered as the place of egg laying. With the exception of a few dates (28 February, 3, 17 March), no significant differences can be found from the results of examinations. The truth of our data is supported by the result of significance calculations; namely, wheat, maize, potato, pea and sunflower as forecrops, provide equally favourable possibilities for egg laying by the female wheat bulb fly. This observation seems to contradict the data obtained by KUPAI (1978), who found that wheat crops sown after a maize forecrop was free from wheat bulb fly. According to our experiences, the wheat bulb flies prefer staying in weedless maize fields, and laying eggs there in the early morning. The opinion that long vegetation forecrops, of row cultivation forming closed stands, provide immunity from wheat bulb flies must, therefore, be modified.

The development of the early wheat shoot fly is linked with the cereals — first of all, with the winter wheat. It, therefore, seemed necessary to settle the question of forecrop in the case of this pest, as well. Thus, parallel to the examination of shoot infestation by wheat bulb fly larvae, 100 sweepings on each occasion were made on the area of investigations (Table 2).

Since the pupae of the early wheat shoot fly hibernate in the soil of fields with winter wheat as forecrop, at the time of the appearance of adults (at the end of February-beginning of March) it is only there that sweeping can be done. At the same time, their colonizing the surrounding winter wheat fields, that had different forecrops, takes place in 7–10 days. Thus,

**Table 1**  
*Infestation of winter wheat shoots  
 by wheat bulb fly larvae after different forecrops*  
 (Velence, 1978)

Date of survey	Percentage of damaged shoots per 10 m				
	winter wheat	maize	potato	pea	sunflower
28 February	0.2	—	0.2	—	—
3 March	3.8	0.1	1.8	0.5	0.4
7	4.1	3.4	3.2	4.2	2.7
10	4.7	3.6	4.1	4.2	3.5
14	4.6	3.7	4.2	2.8	4.4
17	7.6	3.8	8.0	3.6	4.6
21	9.0	6.0	10.5	6.5	7.0
24	9.7	9.1	11.0	11.0	8.5
28	10.5	11.5	13.5	10.5	13.0
31	13.2	13.0	14.0	14.0	14.0
5 April	15.0	12.0	15.2	15.0	15.0
7	16.0	14.1	15.5	16.0	15.5
11	16.5	15.0	15.5	15.5	16.0
14	18.0	16.5	17.0	14.8	18.0
18	17.5	16.8	17.2	16.0	18.5

**Table 2**  
*Number of early wheat shoot fly (Ph. haberlandti Schiner) adults in the insect material obtained  
 by 100 sweepings in winter wheat fields with various forecrops*  
 (Velence, 1978)

Date of sweeping	Forecrops				
	winter wheat	maize	potato	pea	sunflower
28 February	3	—	— +	—	—
3 March	8	—	—	2	—
7	2	—	1	6	4
10	3	3	1	4	1
14	3	1	2	4	3
17	22	23	16	17	9
21	3	8	4	3	6
24	—	—	— +	—	—
28	28	19	17	15	11
31	13	8	4	2	9
5 April	4	1	2	3	4
7	3	2	2	1	2
11	3	—	1	2	2
14	—	—	1	—	—
18	—	—	—	—	—
Total number	95	65	51	59	51

the females may lay eggs in any winter wheat stands regardless of the forecrop. Further, the results of sweeping reveal that the largest number of specimens (95) were found in the field where winter wheat had been the forecrop. In the other fields the adult population was smaller — 51–65 specimens/100 sweepings, depending on the distance and situation. From this we may draw the conclusion that, while the monoculture increases the damage caused by the early wheat shoot fly, fields with other forecrops may also suffer from it; the extent of invasion depends on the distance of the reservoir area. Therefore, by avoiding the neighbourhood of monocultures and fields with winter wheat as forecrop damages caused by the larvae of the early wheat shoot fly can be reduced.

b) Sowing time. Several authors attribute the autumn appearance of harmful species to early sowing. In agreement with earlier investigations (AUFZETZ—JABLONOWSKI 1910, JABLONOWSKI 1916, 1917, 1923, KEREKES 1924, KADOCSA 1943, SÁRINGER 1954, 1958, GOLUBKIN 1963), we found that the early stands greatly suffered from autumn pests, such as the frit fly. As the importance of spring fly damages has recently increased, we felt justified in studying the question of sowing time from this viewpoint, as well.

To test the results of LUTZE—MENDE (1972) in Hungary, we analysed the dependence of damages by wheat bulb fly larvae on the sowing time. In 1978 we evaluated eight winter wheat stands sown at different times in the "Gárdonyi Géza" Co-operative Farm, Agárd. The dates of sowing were: 29 September; 4, 16, 18, 28 October; 3, 6, 9 November 1977.

In the winter wheat fields sown at different times, the extent of infestation by wheat bulb fly larvae was different in spring. Shoot infestation was 11.5 per cent in the stand sown

**Table 3**  
*Surveying results of early wheat shoot fly (Ph. haberlandti Schiner) pupae*  
(Velence, 1979)

Place of survey	Date of survey	Number of pupae per $10 \times 1 \text{ m}^2$										Average, number/ $\text{m}^2$
		1	2	3	4	5	6	7	8	9	10	
I.	Stubble field (3 July)	3	1	4	2	5	2	1	1	2	0	2.2
	After stubble-stripping (6 August)	0	0	1	1	0	2	1	1	0	1	0.7
	Sowing (27 October)	0	1	0	0	1	1	0	0	0	0	0.3
II.	Stubble field (3 July)	2	1	3	0	1	2	1	2	3	2	1.7
	After stubble-stripping (6 August)	1	0	0	0	1	1	0	1	2	0	0.6
	Sowing (27 October)	0	0	0	0	0	0	1	0	0	1	0.2
III.	Stubble field (3 July)	1	0	1	2	1	0	1	1	1	2	1.0
	After stubble-stripping (6 August)	0	0	0	1	1	0	0	1	0	0	0.3
	Sowing (27 October)	0	0	0	0	1	0	0	0	0	0	0.1
IV.	Stubble field (3 July)	2	2	1	3	1	0	1	1	0	1	1.2
	After stubble-stripping (6 August)	1	0	1	1	0	0	0	1	0	0	0.4
	Sowing (27 October)	0	0	1	0	0	0	0	0	1	0	0.2

SD ( $P = 5.0\%$ ) = 0.666    SD ( $P = 1.0\%$ ) = 0.881    SD ( $P = 0.1\%$ ) = 1.134



Table 4

*Average surveying results of early wheat shoot fly  
(Ph. haberlandti Schiner) pupae  
(Velence, 1979)*

Phase of cultivation	Place of survey (field)				Average	
	I	II	III	IV	number/m <sup>2</sup>	%
Stubble field	2.2	1.7	1.0	1.2	1.53	100.0
After stubble-stripping	0.7	0.6	0.3	0.4	0.50	32.6
Crop	0.3	0.2	0.1	0.2	0.20	13.1

at the earliest, and 26 per cent in that sown at the latest date (29 September and 9 November, respectively). The relationship between the time of sowing and the extent of shoot infestation by the wheat bulb fly larvae (Fig. 1) shows a linearly increasing tendency ( $R = 0.967$ , its directional tangent 0.355). This can be explained by the fact that stands sown earlier become stronger before winter sets in, overwinter in a more resistant physiological condition and, in spring, fewer shoots of them fall victim to the larvae of chloropid flies. Abundantly tillering wheat varieties can replace the shoots destroyed by flies; but wheat plants sown late, and consequently underdeveloped, are not capable of doing so. Keeping to the optimum time of sowing considerably reduces the spring damages by flies.

c) Effect of implements on the number of population. LUTZE—MENDE (1972) carried out thorough studies on the effect of soil cultivation on the population number of the wheat bulb fly. In the course of their investigations, they found that the number of harmful larvae was not even reduced when the eggs had been worked into a soil depth of 15–25 cm. During our observations made over several years (1974–1981), this fact also proved true under Hungarian conditions. Damages caused by the larvae of the wheat bulb fly cannot be decreased merely by soil cultivation methods.

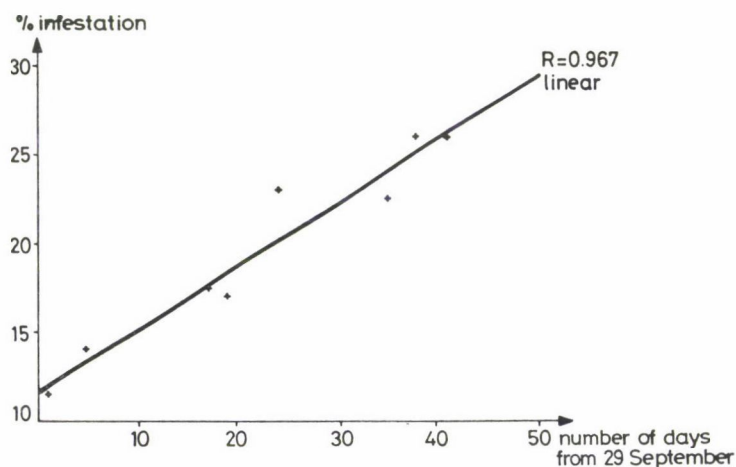


Fig. 1. Extent of wheat bulb fly infestation as a function of sowing time

Table 5

*Percentage of emergence of  
early wheat shoot fly adults  
(Ph. haberlandti Schiner)  
from pupae at various depths  
of soil  
(Velence, 1980)*

Depth below soil surface (cm)	Adult emergence (%)
2	87.5
10	22.5
18	0
26	0

At the time of the soil cultivation operations, the pupae of the early wheat shoot fly are in the upper several centimeters of the soil. We thought it therefore important to study the mechanical effect of implements on the pupae, and the survival of pupae that go deeper in the course of ploughing. In 1979 we made surveys of early wheat shoot fly pupae in four winter wheat monocultures situated close to one another in the following phases of cultivation: (Tables 3—4)

- in stubble field (3 July) with the original condition of soil;
- after stubble-stripping (6 August);
- in newly sown stand (27 October).

In the course of ploughing, a considerable percentage of the pupae left intact by the implements gets into deeper soil; so, another question we had to answer was whether the pupae in the different soil horizons were able to develop into viable adults. To this end we set up an isolator experiment. In four replications, 160 early wheat shoot fly pupae, that had hibernated at depths of 2, 10, 18 and 26 cm, respectively, were selected. The pupae were put into plastic cylinders and placed in sand to the same four depths as above. The cylinders were covered at the top with close-woven nets made of synthetic material and put into the sand at the four appropriate depths. The evaluation was made in spring when the adults swarmed, by determining the percentage that emerged (Table 5). Besides the previous injuries caused by implements, the figures of Table 5 indicate further losses; namely, adults developed only from 22.5 per cent of the otherwise viable pupae at the depth of 10 cm. From deeper layers (18–26 cm) no adults emerged. Thus, even when uninjured by implements used in ploughing, most of the pupae would suffocate when buried deeper than 10 cm in the soil. Deep ploughing is therefore an efficient method of controlling the early wheat shoot fly.

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#### References

- ANONYMOUS (1928): A gabonalegyekről (The chloropid flies). *Növényvédelem*, **8**, 160.  
 ANONYMOUS (1971): Sie trifft das Herz der Pflanze. Die Gefährlichkeit der Brachfliege in Getreide wird oft unterschätzt. *Pflanzenschutz-Kurier Bayer*, **16**, 62–63.  
 AUFZETZ, J.—JABLONOWSKI, J. (1910): Légykárók és a késő őszi vetések (Fly damages and late autumn sowing). *Köztelek*, **20**, 877.

- BOLLOW, H. (1960): Über ein diesjähriges Auftreten der Brachfliege (*Phorbia coarctata* Fall.) in Bayern. Pflanzenschutz, **12**, 139–142.
- BUHL, C. (1963): Dipteren als Getreideschädlinge unter Berücksichtigung der Fruchtfolge. Mitt. biol. Bundesanst. Land- u. Forstwirtsch., **108**, 158–162.
- DARVAS, B.—D. DRASKOVITS, A.—PAPP, L.—HEGEDÜS, I.—LESZNYÁK, M.—SZEŐKE, K. (1981): Az őszi búza légykártevői I. Kártétfelmérés, rajzásdinamikai vizsgálatok (Fly pests of winter wheat I. Assessment of damages, examinations of swarming dynamics). Növényvédelem, **17**, 96–109.
- FABER, W. (1968): Brachfliege [*Phorbia*(*Leptohylemyia*)*coarctata* Fall.]. Bundesanstalt für Pflanzenschutz, Wien. Flugblatt Nr. 133.
- FRANK, A. (1900): Beschädigungen des Wintergetreides durch die Getreide — Blumenfliege *Hylemyia coarctata* (Fall.). Arb. biol. Abth. Land- u. Forstwirtsch., **1**, 265–267.
- GEMMIL, J. F. (1923): Wheat bulb disease. Scottish J. Agric., **6**, 192–196.
- GOLUBKIN, V. G. (1963): A korai vetések és a fritlégy (Early sowing and the frit fly). Zasc. Vred. Bolezn., Moscow, 8/10, 17.
- GOUGH, H. C. (1953): The problem of the wheat bulb fly. J. Ministry Agric., **60**, 315–320.
- GOUGH, H. C. (1957): Wheat bulb fly. Biological and agricultural problems, Ann. appl. Biol., **45**, 384–385.
- GYÖRFFY, J. (1927): Mikor szántunk ki a gabonalegyek által megtámadott vetést (When to plough out stands infested by chloropid flies). Növényvédelem, **3**, 11.
- JABLONOWSKI, J. (1901): Védekezés a hesszeni légy ellen (Protection against Hessian fly). Köztelek, **11**, 1169–1170.
- JABLONOWSKI, J. (1916): Mi fenyegeti a jövő évi vetésünket? (Hesszeni légy) [What threatens our next year crop? (Hessian fly)]. Magyar földműves, **7**, 267–269.
- JABLONOWSKI, J. (1917): Az őszi vetés és a hesszeni légy (Autumn sowing and the Hessian fly). Debreceni Gazdasági Lapok, **20**, 21–22.
- JABLONOWSKI, J. (1923): Hesszeni legyes vetés — sárguló vetés (Hessian fly infested crop — yellowing crop). Köztelek, **33**, 1111–1112.
- JABLONOWSKI, J. (1927): Az őszi búzavetés rovarkártevői (Észrevételek egy hozzászóláshoz) [Insect pests of autumn wheat crops (Contribution to a debate)]. Köztelek, **37**, 588–589.
- JABLONOWSKI, J. (1928): Vetésforgó s a kártékony rovarok. Toldalék Westsik V. közleményéhez (Crop rotation and the noxious insects. Addition to V. Westsik's publication). Köztelek, **38**, 1658–1661.
- JERMY, T. (1953a): A fekete búzalegyek (*Phorbia securis* Tiensuu, *Phorbia penicillifera* n. sp.), (Diptera, Anthomyiidae) [Wheat shoot flies (*Phorbia securis* Tiensuu, *Phorbia penicillifera* n. sp.), (Diptera, Anthomyiidae)]. Növényvédelmi Kutató Intézet Évkönyve, **6**, 55–86.
- JERMY, T. (1953b): Beitrag zur Kenntnis der schwarzen Getreideblumenfliegen (*Phorbia securis* Tiensuu, *Phorbia penicillifera* Jermy: Diptera, Anthomyiidae). Acta Agron. Hung., **3**, 225–255.
- JERMY, T.—SZELÉNYI, G. (1958): Az őszi búza állattársulásai (Animal associations in winter wheat). Állattani Közlemények, **46**, 229–241.
- KADOCSA, GY. (1923): Gabonalegyek elleni védekezés főbb szabályai (Major rules in the control of chloropid flies). Gazdasági Lapok, **75**, 51.
- KADOCSA, GY. (1943): Védekezés a gabonalegyek ellen (Control of chloropid flies). Pátria Irodalmi Vállalat és Nyomdai Részvénytársaság, Budapest, 20.
- KEREKES, F. (1924): Gabonaféléket pusztító legyek kártételei és az ellenük való védekezés (Fly pests of cereals and possibilities of control). Magyar Rósa, **2**, 5–6.
- KLEINE, (1918): Die Getreideblumenfliege (*Hylemyia coarctata*) Diesjährige Beobachtungen in Pommern. Z. angew. Entomol., **4**, 16–24.
- KUPAI, J. (1972): A tavaszi gabonalegy kárról (Damage done by the early wheat shoot fly). Magyar Mezőgazdaság, **27/20**, 10–17.
- KUPAI, J. (1978): Ugárlégy (*Leptohylemyia coarctata* Fallén) Magyarországon [Wheat bulb fly (*Leptohylemyia coarctata* Fallén) in Hungary]. Doctor's thesis, Keszthelyi Agrártudományi Egyetem, Keszthely, 91.
- KÜKEDI, E. (1975): Az ugárlégyről (*Phorbia coarctata* Fall.) [The wheat bulb fly (*Phorbia coarctata* Fall.)]. Növényvédelem, **11**, 255–262.
- LUTZE, G.—MENDE, F. (1972): Biologie und Bekämpfung der Brachfliege (*Leptohylemyia coarctata* Fallén). Dissertation dem Wissenschaftlichen Rat der Martin Luther Universität Halle—Wittenberg. Wittenberg.
- MASKELL, F. E.—DAVIS, M. E. (1974): Bekämpfungsmaßnahmen mit Folimat gegen die Brachfliege (*Phorbia coarctata* Fall.). Pflanzenschutz-Nachrichten Bayer, **27**, 295–311.



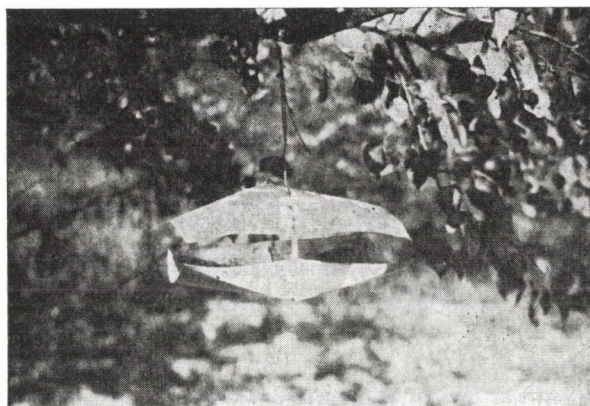
- MENDE, F. (1974): Grundlagen zur Befallsprognose der Bruchfliege (*Leptohylemyia coarctata* Fallén). Symposium mit internationaler Beteiligung zur Schäderregerüberwachung in der industriemäßigen Getreideproduktion. Halle. Manuskriptdruck der Vorträge Teil I., 95–103.
- MEZEY, GY. (1899): A hesszeni légy és a rothadó búza — meg egy kis helyreigazítás (The Hessian fly and the decaying wheat — and some correction). Köztelek, **9**, 1197.
- MEYER, W. (1976): Untersuchungen zum Auftreten der Bruchfliege (*Leptohylemyia coarctata* Fall.) und Ergebnisse von Bekämpfungsversuchen bei Winterweizen. Mitteilungen, **24**, 189–202.
- MIEGROET, M. (1950): Bijdrage tot de kennis van de biologie, de ecologie en de economische betekenis van *Hylemyia coarctata* Fall. Meded. Land. Hogesch, Gent, **15**, 270–303.
- PERJU, T.—PÉTERFY, F. (1970): Grey fly [*Phorbia*(*Leptohylemyia*)*coarctata* Fall.] injuries to cereal crops in Transylvania. Probleme Agricole, Kolozsvár, Romania, **22**, 42–48.
- POKORNY, J. (1927): Drovkridli skudei obilnin. Stud. Inf. Ochr. Rost, **5**, 54.
- RAPAICS, R. (1914): Védekezés a fritlégy ellen (Control of frit fly). Gazdasági Lapok, **66**, 10.
- RECAMIER, A. (1964): Observations sur l'extension des dégats de la mouche grise du blé au Centre National d'Experimentation et étude de certains moyens de lutte. Comptes rendus des séances de l'Académie d'Agriculture de France, **50**, 773–779.
- ROSTRUP, S. (1911): Lebensweise der *Hylemyia coarctata* in Denmark. Z. Pflanzenkrankh. (Pflanzenpathol.) Pflanzenschutz, **21**, 386–387.
- SÁRINGER, GY. (1950): A gabonalegyek országos elterjedésének vizsgálata 1950-ben (Study of the nation-wide distribution of chloropid flies in 1950). Agrártudomány, **2**, 476–483.
- SÁRINGER, GY. (1954): Miért sárgulnak az őszi vetések (Why do the autumn crops turn yellow). Magyar Mezőgazdaság, **9/24**, 8.
- SÁRINGER, GY. (1958): Hozzászólás Pusztai A. "Az . . . őszi búza kelésére . . ." c. cikkéhez növényvédelmi szempontból (Contribution of plant protection aspects to A. Pusztai's paper: ". . . on the emergence of winter wheat"). Magyar Mezőgazdaság, **13/17**, 6.
- SZEŐKE, K. (1981): Az őszi búzát károsító viráglegyek (*Anthomyiidae*) életmódja és védekezés-technológia [Ethology of the flower fly (*Anthomyiidae*) pests of winter wheat, and technology of control]. Doctor's dissertation, Keszthelyi Agrártudományi Egyetem, Mezőgazdaságtudományi Kar, Mosonmagyaróvár, 146.

#### PHEROMONE BAITED TRAPS: EFFECT OF TRAP TYPE AND PHEROMONE RATE ON TRAP EFFICIENCY; FIELD TRIALS ON CODLING MOTH (*CYDIA POMONELLA* L.) IN HUNGARY

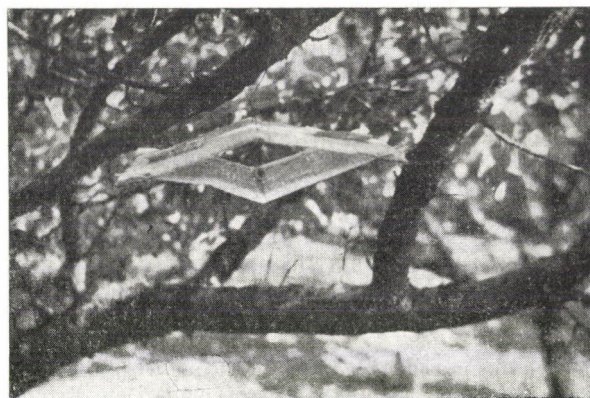
Trials were carried out in abandoned apple orchards with a high population level, and in commercial orchards with a low population level. Comparisons were made between three types of traps, and pheromone rates of 2 mg, 1 mg, 0.5 mg and 0.25 mg were tested. The results revealed that among those tested, the most suitable trap for codling moth monitoring was the Pherocon 1C trap. In the pheromone trials the results showed that a rate of 1 mg in a Zocon cap is more efficient than 1 mg in a Reanal cap in districts with high populations, but both caps with the same pheromone rate are equally attractive in districts with low populations. Fresh rubber tube septa having initial charges of 1 mg or 0.5 mg attracted a numerically high number and showed a significant difference compared to the other rates. A rate of 2 mg was significantly less attractive than the other rates used in the trials.

The sex pheromone of the codling moth (*Cydia pomonella* L., Lepidoptera: Tortricidae) has been determined by ROELOFS *et al.* (1971) as (E,E)-8,10 dodecadien-1-1-ol. With the development of a method for synthesizing this compound in a pure form, field researchers in the field of population estimation have been granted the opportunity to develop a new method of population estimation using traps baited with this compound. Previous workers (CLUVER—BARNES 1977) stated that the trap type and pheromone rate influenced the sex pheromone trap catch. In these studies the effect of trap type and different pheromone rates on sex pheromone trap efficiency have been tested.

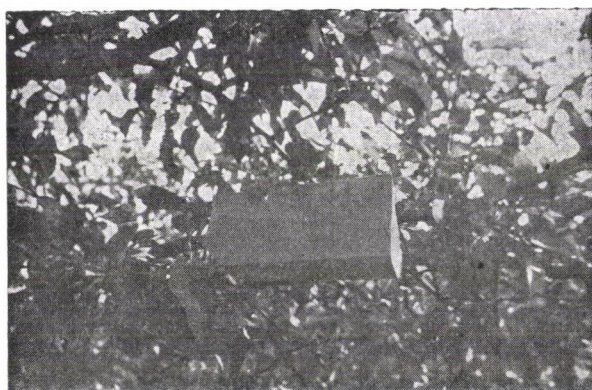
*Orchards:* The trials were carried out at the following locations: 1. An abandoned orchard at Kamaraerdő in the environs of Budapest (Research Station of the University of Horticulture) which had a high population of codling moth; it contained more than one native variety of apple, planted more than thirty years ago, and had not received an organized programme of spraying for a long time. 2. A commercial orchard (model farm) belonging to the University of Horticulture that is also an Experimental Research orchard, cultivated with Jonathan, Golden Delicious and Starking. This orchard is located at Laki-hegy. 3. A house



*Fig. 1. Pherocon 1C trap*



*Fig. 2. Reamol trap*



*Fig. 3. Sector-like trap*



garden at Mátyás-hegy, in the environs of Budapest, planted with different apple varieties. 4. The orchards of the Izsák Cooperative Farm and the Kis-fái Model Farm (Research Station of the Horticulture College at Kecskemét) which are commercial orchards established more than ten years ago and planted with the major apple varieties.

*Trap designs:* comparisons were made between three types, namely:

1. Pherocon 1C trap (Zoecon Corporation Palo Alto, CA 94304) which is a rectangular shape (Fig. 1), consisting of weatherproofed paperboard upper and lower sections, the lower being coated with adhesive on its upper surface. The two sections (top and bottom) are held together by a wire hanger and spacer.

2. Reamol trap (Hungarian product manufactured by the Reanal Corporation, 1147 Budapest, Hungary) which is lozenge-shaped (Fig. 2) and consists of a top and bottom made of weatherproofed synthetic material, connected together by means of four projections on the bottom (two at each narrow side) inserted into four oblique openings in the top corners. The bottom is covered with adhesive on its upper surface.

3. Sectar-like trap which is a cardboard trap, triangular in shape when in use (Fig. 3). The sticky surface is a replaceable liner, replaced whenever necessary (usually after 4 weeks). The trap was designed by Dr. H. Arn (Research Station, Wädenswil, Switzerland) and was used from 10 July 1979, when it was acquired, until the end of the trials.

*Pheromone:* The pheromone used came from two sources: the standard caps made by Zoecon and Reanal, which are rubber caps impregnated with 1 mg Codlemone, and a rubber tube impregnated with Codlemone (brought from Dr. P. J. Charmillot, Research Station, Changins, Switzerland).

*Assessment.* The catches at Kamaraerdő and Laki-hegy were recorded weekly, and those at Mátyás-hegy, Izsák and Kis-fái daily, but the catches at these locations were also transformed into weekly catches. The moths were removed from the traps after identification and recording.

### 1. Trap design and efficiency

#### *At Kamaraerdő (area of high population)*

The trial period lasted 23 weeks (from 9. 5. 79 to 16. 10. 79). In the first 9 weeks two Pherocon 1C traps were used, each baited with one Zoecon cap, and one Reamol trap baited with one Reanal cap. The results for this period (Table 1a) indicated that there was a significant difference between the two types. In the second period (after 10. 7. 79), which lasted 14 weeks, in addition to the afore-mentioned traps a sectar-like trap baited with a section of rubber tube containing 1 mg codlemone was also tested. The results for this period (Table 1b) revealed that there were significant differences between the three types in catching efficiency.

#### *At Laki-hegy (area of low population)*

The period of study, trap types, and pheromone sources were exactly the same as for Kamaraerdő. The trapping results are presented in Table 2. There was a significant difference between the means of catches for the different types.

#### *At Izsák and Kis-fái (areas of low population)*

The study period lasted 17 weeks (from 9. 5. 79 to 4. 9. 79). At Izsák one Pherocon 1C trap baited with one Zoecon cap and one Reamol trap baited with one Reanal cap were used and at Kis-fái one Reamol trap baited with one Reanal cap. The results are given in Table 3. There was no significant difference between the two types at Izsák, but there was a significant difference between the Reamol trap at Kis-fái and both the Pherocon 1C trap and the Reamol trap at Izsák.

The trapping results showed that catch size is influenced by trap shape and dimensions. There was a significant increase in the number of male moths captured in traps having the largest basal area and greatest width. The effective trapping area was approx. 504 cm<sup>2</sup> for both the Pherocon 1C trap (Fig. 4) and the Reamol trap (Fig. 5), while this area was approx. 153 cm<sup>2</sup> in the sectar-like trap (Fig. 6). The high efficiency of the Pherocon 1C trap compared to that of the Reamol trap is, in our opinion, due to the difference between the two types in volume and construction. The volume of the Pherocon 1C trap is approx. 3310 cm<sup>3</sup> and it has a side spacer 5 cm long, while the Reamol trap has a volume of approx. 1814 cm<sup>3</sup> and it has no side spacer. As regards the state of the adhesive surface, in the Pherocon 1C trap the catch-



Table 1

*Number of male codling moths captured in sex pheromone traps of various designs at Kamaraerdő, 1979*

Date	Trap designs			
	Pherocon 1C		Reamol No. 3	Sectar-like No. 4
	No. 1	No. 2		
<i>a)</i>				
15. 5.	20	18	2	—
22. 5.	128	134	2	—
29. 5.	61	79	22	—
6. 6.	17	22	6	—
13. 6.	10	19	19	—
20. 6.	17	21	3	—
26. 6.	18	13	22	—
3. 7.	95	112	1	—
10. 7.	52	41	5	—
Total	418	469	82	
Mean of trap catches per week	46.4 <sup>ab</sup>	52.1 <sup>ab</sup>	9.1 <sup>a</sup>	
<i>b)</i>				
17. 7.	38	98	2	51
24. 7.	32	69	0	16
31. 7.	5	50	21	18
7. 8.	3	52	0	23
14. 8.	1	30	0	12
21. 8.	98	19	11	11
28. 8.	47	16	0	2
4. 9.	3	4	0	0
12. 9.	0	0	0	0
18. 9.	5	0	0	0
25. 9.	1	0	0	0
2. 10.	1	1	0	0
9. 10.	0	0	0	0
16. 10.	0	0	0	0
Total	234	339	34	133
Mean of trap catches per week	16.7 <sup>ab</sup>	24.8 <sup>ab</sup>	2.4 <sup>a</sup>	9.5 <sup>a</sup>

(—) Means the trap was not hung until after this date.

Data analysed by LSD test; means with different letters are significantly different at the 5% level.

ing surface is coated with a sufficient quantity of glue, which is of good viscosity and stickiness and is not affected by weather factors; but in the Reamol trap the adhesive material is of poor quality and insufficient quantity. For the sectar-like trap, the low number of catches is due to the small basal area (approx. 153 cm<sup>3</sup>), small volume (approx. 590 cm<sup>3</sup>) and the absence of a side spacer. The results do not agree with those of ROELOFS *et al.* (1973), which indicated that a decrease in the height of the trap opening (types with the bottom and top sections closed), as in the Reamol and sectar-like traps, led to an increase in trap catches. Nor do the results agree with those of LEWIS—MACAULAY (1976) which showed that the trap catch increased with an elongation of the pheromone odour plume, as found in closed (top and bot-

Table 2

*Number of male codling moths captured in sex pheromone traps of various designs at Laki-hegy, 1979*

Date	Trap designs			
	Pherotrap 1C		Reamol No. 3	Sectar-like No. 4
	No. 1	No. 2		
a) First period				
16. 5.	14	9	2	—
23. 5.	54	35	1	—
30. 5.	8	13	6	—
7. 6.	12	13	2	—
12. 6.	3	1	0	—
20. 6.	1	3	0	—
26. 6.	0	4	0	—
3. 7.	11	0	1	—
10. 7.	31	7	8	—
Total	134	85	20	—
Mean of trap catches per week	14.9 <sup>ab</sup>	9.4 <sup>a</sup>	2.2	—
b) Second period				
17. 7.	7	8	1	3
24. 7.	8	3	0	4
31. 7.	6	0	2	5
7. 8.	8	2	2	7
14. 8.	1	0	0	0
21. 8.	11	6	0	1
28. 8.	4	1	0	0
4. 9.	0	1	0	0
12. 9.	2	1	0	0
18. 9.	1	1	0	0
25. 9.	1	0	0	0
3. 10.	0	0	0	0
9. 10.	0	0	0	0
16. 10.	0	0	0	0
Total	49	23	5	20
Mean of trap catches per week	3.5 <sup>ab</sup>	1.6 <sup>a</sup>	0.36 <sup>a</sup>	1.4 <sup>a</sup>

(—) Means the trap was not hung until after this date.

Data analysed by LSD test; means with different letters are significantly different at the 5% level.

tom) trap types. The present results suggested that an omnidirectional trap (top and bottom not closed) with a pheromone odour plume evaporating radially (Pherocon 1C trap) gave the best catches.

## 2. Pheromone rate, dispenser type, and sex trap catch

### *At Kamaraerdő (area of high population)*

The trials lasted 23 weeks (from 8. 5. 79 to 16. 10. 79). In the first 9 weeks (up to 10. 7. 79) comparison were made between rates of 1 mg (Zoecon) in a Pherocon 1C trap, 1 mg (Reanal) in a Reamol trap, 2 mg (Reanal) in a Reamol trap and 1 mg (Reanal) in a Pherocon 1C trap.

**Table 3**

*Number of male codling moths captured in sex pheromone traps of various designs at Izsák and Kisfái, 1979*

Date	Trap design		
	at Izsák		at Kisfái Reamol No. 3
	Pherocon 1C No. 1	Reamol No. 2	
17. 5.	0	0	0
24. 5.	13	1	6
31. 5.	20	7	0
7. 6.	7	12	2
14. 6.	7	5	0
21. 6.	5	0	0
28. 6.	6	1	1
4. 7.	2	4	0
11. 7.	37	16	0
18. 7.	3	1	0
25. 7.	1	7	0
31. 7.	4	6	1
7. 8.	21	10	1
14. 8.	2	25	0
28. 8.	1	6	1
4. 9.	2	5	0
Total	133	108	13
Mean of trap catches per week	7.8 <sup>ab</sup>	6.35 <sup>ab</sup>	0.76 <sup>a</sup>

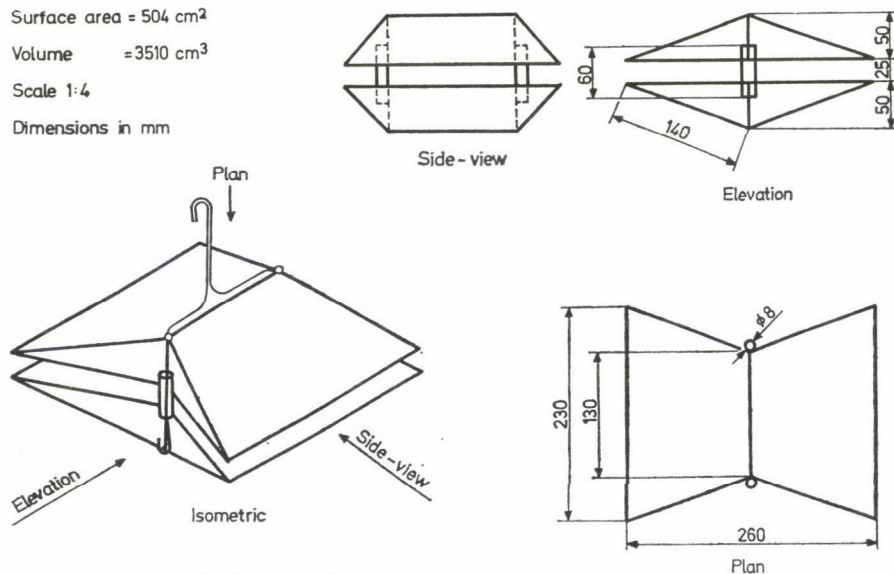
Data analysed by LSD test; means with different letters are significantly different at the 5% level.

Surface area = 504 cm<sup>2</sup>

Volume = 3510 cm<sup>3</sup>

Scale 1:4

Dimensions in mm



PHEROCON 1C TRAP

Fig. 4. Pherocon 1C trap; dimensions and surface area



Surface area =  $504 \text{ cm}^2$

Volume =  $1814 \text{ cm}^3$

Scale 1:4

Dimensions in mm

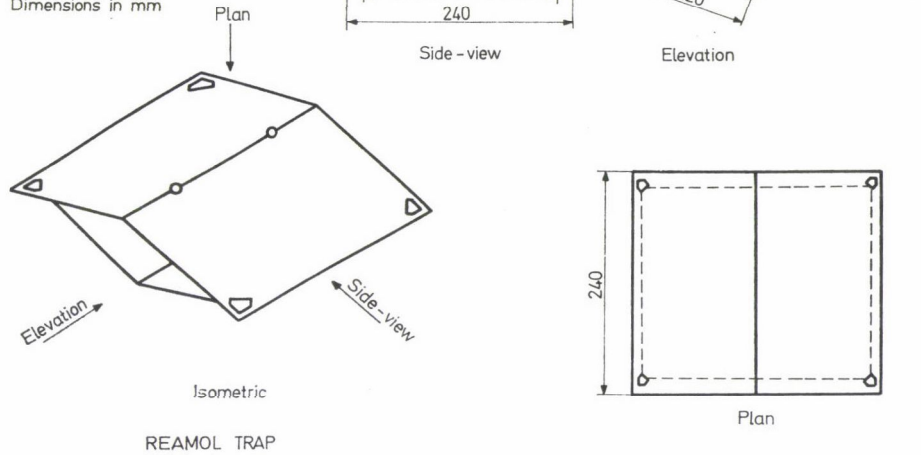


Fig. 5. Reamol trap; dimensions and surface area

Scale 1:2

Dimensions in mm

Surface area =  $153 \text{ cm}^2$

Volume =  $590 \text{ cm}^3$

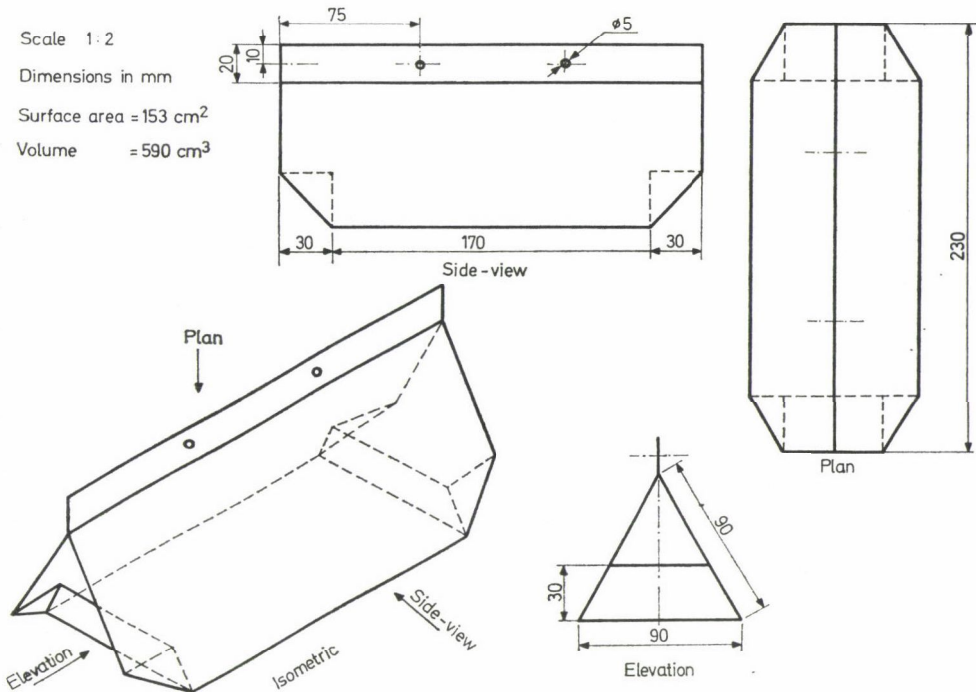


Fig. 6. Sectar-like trap; dimensions and surface area

Table 4

Number of male codling moths captured in sex pheromone traps with different pheromone concentrations and different pheromone releasers at Kama-raerdő, 1979

		Trap design pheromone concentrations and releaser							
Date		Pherocon 1C baited with 1 mg Codlemone in a Zoecon rubber cap	Reamol baited with 1 mg Codlemone in a Reanal rubber cap	Reamol baited with 2 mg Codlemone in a Reanal rubber cap	Pherocon 1C baited with 1 mg Codlemone in a Reanal rubber cap	Pherocon 1C baited with 1 mg Codlemone in a Swiss rubber tube	Pherocon 1C baited with 0.5 mg Codlemone in a Swiss rubber tube	Pherocon 1C baited with 0.25 mg Codlemone in a Swiss rubber tube	Sectar-like baited with 1 mg Codlemone in a Swiss rubber tube
a)	15. 5.	20	2	3	17	—	—	—	—
	22. 5.	128	2	1	31	—	—	—	—
	29. 5.	61	22	0	19	—	—	—	—
	6. 6.	17	6	0	14	—	—	—	—
	17. 6.	10	19	10	16	—	—	—	—
	20. 6.	17	3	0	24	—	—	—	—
	26. 6.	18	22	5	18	—	—	—	—
	3. 7.	95	1	0	41	—	—	—	—
	10. 7.	52	5	4	13	—	—	—	—
Total		418	82	23	189	—	—	—	—
Mean of trap catches per week		46.4 <sup>ab</sup>	9.1 <sup>a</sup>	2.6 <sup>a</sup>	21 <sup>a</sup>	—	—	—	—
b)	17. 7.	38	2	2	20	79	61	15	51
	24. 7.	32	0	8	18	183	59	9	16
	31. 7.	5	21	8	17	59	28	8	18
	7. 8.	3	0	1	42	39	11	0	23
	14. 8.	1	0	0	22	18	12	9	12
	21. 8.	98	11	5	15	18	96	138	11
	28. 8.	47	0	1	2	3	8	17	2
	4. 9.	3	0	0	2	4	3	11	0
	12. 9.	0	0	0	1	0	2	3	0
	18. 9.	5	0	0	3	0	0	0	0
	25. 9.	1	0	0	0	0	0	1	0
	2. 10.	1	0	0	0	0	1	0	0
	9. 10.	0	0	0	0	0	0	0	0
	16. 10.	0	0	0	0	0	0	0	0
Total		234	34	25	142	403	281	211	133
Means of trap catches per week		16.7 <sup>a</sup>	2.4 <sup>a</sup>	1.8 <sup>a</sup>	10.1 <sup>a</sup>	28.8 <sup>ab</sup>	20.07 <sup>a</sup>	15.07 <sup>a</sup>	9.5 <sup>a</sup>

(—) Means the traps were not hung until after this date.

Data analysed by LSD test; means with different letters are significantly different at the 5% level.

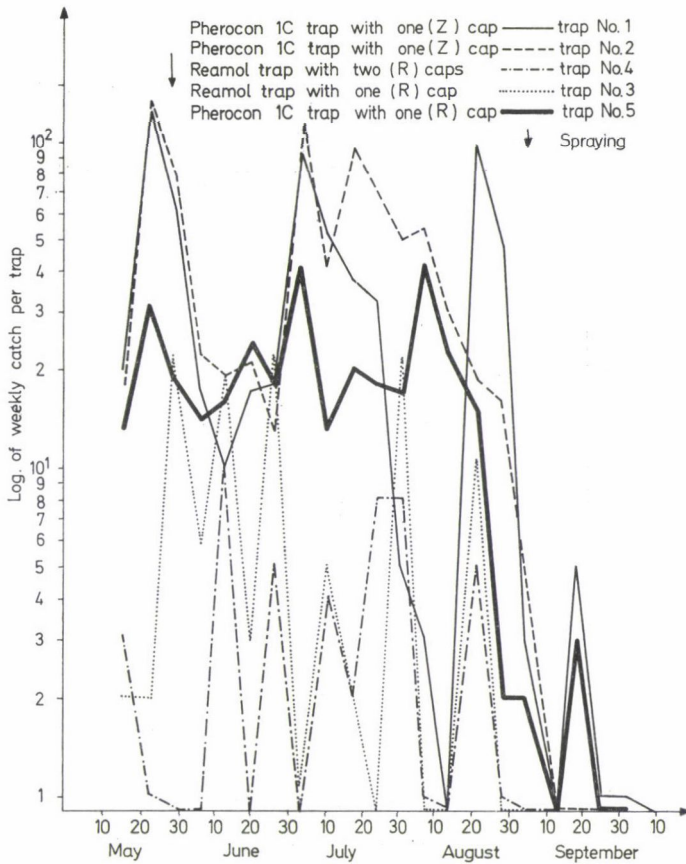


Fig. 7. The codling moth: Seasonal pheromone trapping at Kamaraerdő, 1979

In the following 14 weeks (from 10. 7. 79 until the traps were removed), besides the aforementioned rates, sections of rubber tube impregnated with Codlemone (brought from Switzerland) were also tested. The tubes contained 1 mg, 0.5 mg or 0.25 mg in the Pherocon 1C trap and 1 mg in the sector-like trap. The Zoecon caps were replaced every 6 weeks, the Reanal caps every two weeks, and the rubber tubes every 6 weeks. The weekly catches are presented numerically in Table 4 and graphically in Figs 7 and 8. The results indicated that there was a significant difference between the means of total trap catches per week in the first 9 weeks, and that Zoecon caps with 1 mg were the most effective. In the second period, the results showed that there was a significant difference between the catch means achieved with 1 mg and 0.5 mg in a rubber tube dispenser used in a Pherocon 1C trap compared to other rates and releasers. A rate of 1 mg in a Toecon cap caught a high number, though it was less efficient than the 1 mg and 0.5 mg rates in rubber tube sections; this is due to the fact that the rubber tube was fresh, while the Zoecon cap was more than a year old. A rate of 2 mg gave a low attraction level, due to the flight response inhibition caused by high pheromone rates.

*At Mátyáshegy (house garden, area of high population)*

Comparisons were made between a pheromone rate of 1 mg supplied by one Zoecon cap baited in a Pherocon 1C trap, and a pheromone rate of 2 mg from two Reanal caps baited in a Reamol trap. The Zoecon caps were replaced every 6 weeks, while the two Reanal caps



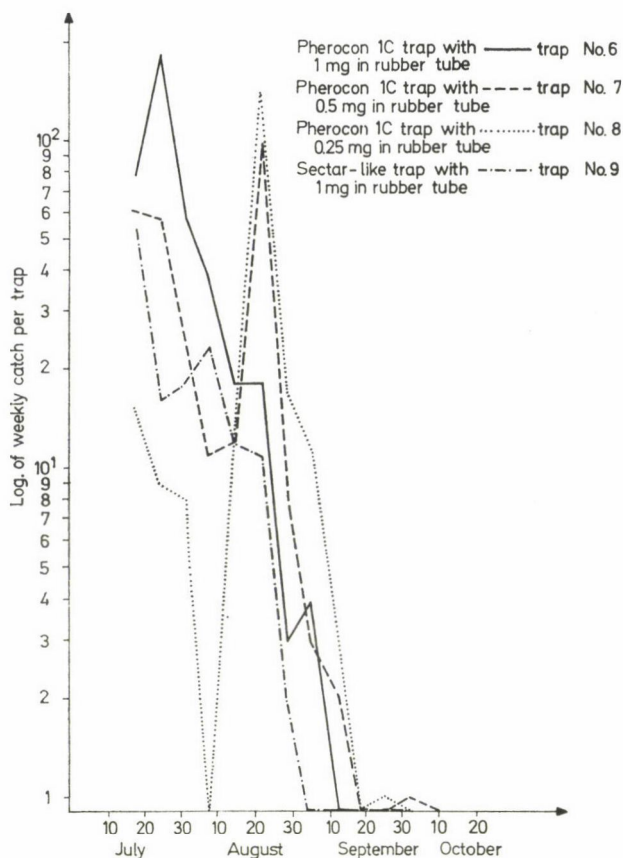


Fig. 8. The codling moth: Pheromone trapping at Kamaraerdő, 1979

were not replaced throughout the trial period. The trial period lasted 16 weeks (from 25. 5. 79 to 25. 9. 79).

The weekly catches are presented numerically in Table 5 and graphically in Fig. 9. There was a significant difference between the means of total catch per week for the two pheromone rates. It was observed that the Reamol trap with 2 mg showed a low attraction efficiency after the first week, and it did not catch any male moths after the 4th week (13. 6. 79). Here the inhibition effect of a high pheromone rate on the flight response is very clearly seen.

#### *At Laki-hegy (area of low population)*

The study period lasted 23 weeks (from 9. 5. 79 to 16. 10. 79). The weekly catches are given numerically in Table 6 and graphically in Figs 10 and 11. Different dispensers using different sources were tested. In the first 9 weeks (up to 10. 7. 79) one cap in a Pherocon 1C trap and one Reanal cap in a Reamol trap were used. The Zoecon caps were replaced every 6 weeks and the Reanal caps every two weeks. The results for this period showed that there was no significant difference between the two types of releaser. In the second period (after 10. 7. 79), which lasted 14 weeks, besides the afore-mentioned releasers, two sections of rub-

Table 5

*Number of male codling moths captured in sex pheromone traps of various designs, different pheromone rates and different pheromone releasers at Mátyáshegy, 1979*

Date	Trap designs, pheromone rates and releaser	
	Pherocon 1C baited with 1 mg Codlemone in a Zoecon rubber cap	Reamol baited with 2 mg Codlemone in a Reanal rubber cap
26. 5.	43	3
1. 6.	36	3
7. 6.	70	10
13. 6.	34	2
20. 6.	5	0
26. 6.	45	0
1. 7.	24	0
6. 7.	18	0
17. 7.	66	0
25. 7.	20	0
30. 7.	27	0
4. 8.	19	0
10. 8.	15	0
15. 8.	23	0
20. 8.	6	0
25. 8.	11	0
31. 8.	5	0
25. 9.	66	0
Total	533	18
Mean of trap catches per week	32.7 <sup>ab</sup>	1.0 <sup>a</sup>

Data analysed by LSD test; means with different letters are significantly different at the 5% level.

ber tube each impregnated with 1 mg Codlemone (fresh preparation, brought from Switzerland) were used, one piece baited in a Pherocon 1C trap and the other in a sector-like trap. The rubber pieces were replaced every 6 weeks. The results showed that there was no significant difference between the means of catches, in spite of the high number captured in Pherocon 1C traps baited with a Zoecon cap or a piece of rubber tube impregnated with 1 mg Codlemone. This signified that there is no difference between dispensation from a closed cap or an open rubber tube.

From the results obtained it can be concluded that the type of trap, the shape and the surface area affect the size of male codling moth catches. It can also be stated that the Pherocon 1C trap is a convenient type in relation to the size of catch compared with other types tested. In monitoring studies, the use of Pherocon 1C or Reamol traps showed the same trend for the insect population, so both types are a useful tool in this respect.

In relation to the pheromone rate, an amount of more than 1 mg Codlemone per trap was less efficient in attracting moths. This can be attributed to the inhibiting effect of pheromone at higher rates. The age of the caps can affect the size of catch, but the type of dispenser has no effect on the catch when the same amount of pheromone is used.

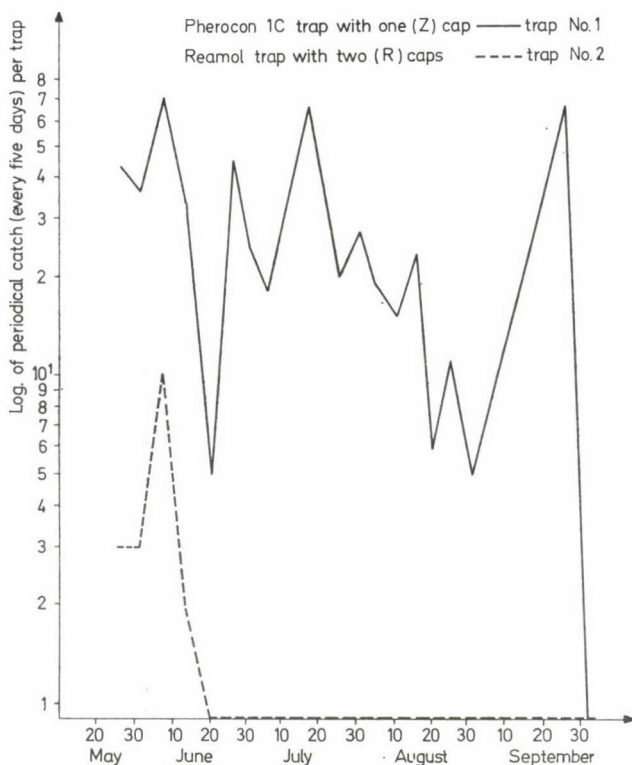


Fig. 9. The codling moth: Seasonal pheromone trapping at Mátyáshegy, 1979

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Prepared at the Plant Protection Department, University of Horticulture, Budapest, Hungary

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#### References

- CLUVER, D. J.—BARNES, M. M. (1977): Contribution to the use of the synthetic pheromone in monitoring codling moth populations. *J. Econ. Entomol.*, **70**, 489–492.
- LEWIS, T.—MACAULAY, E. D. M. (1976): Design and elevation of sex-attractant traps for pea moth, *Cydia nigricana* (Steph.) and the effect of plume shape on catches. *Ecol. Ent.*, **1**, 175–187.
- ROELOFS, W. L.—COMEAU, A.—HILL, A.—MILICEVIS, G. (1971): Sex attractant of the codling moth: characterization with electroantennograms technique. *Science*, **174**, 297–299.
- ROELOFS, W. L.—GRADE, R. T.—TETTE, J. P. (1973): Oriental fruit moth attractant synergists. *Environ. Ent.*, **2**, 252–254.



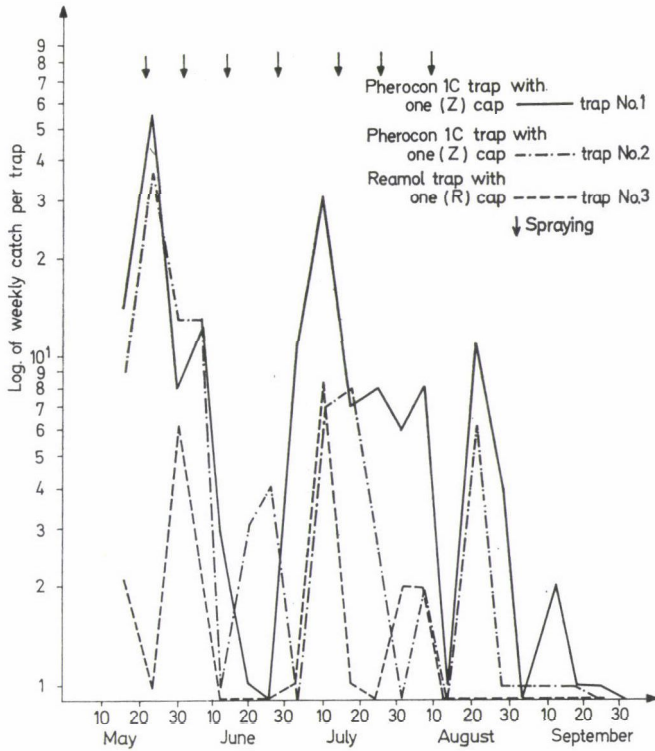


Fig. 10. The codling moth: Seasonal pheromone trapping at Laki-hegy, 1979

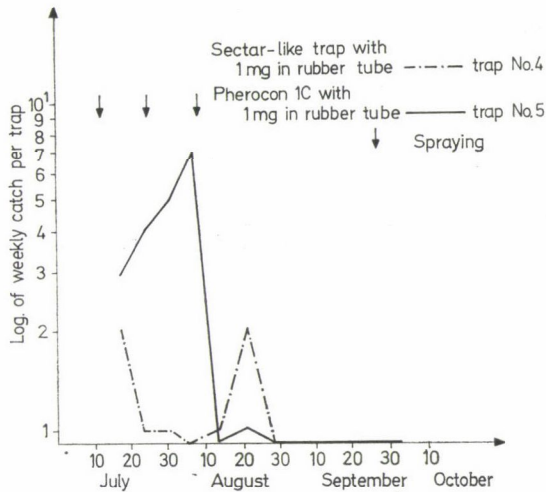


Fig. 11. The codling moth: Pheromone trapping at Laki-hegy, 1979

Table 6

*Number of male codling moths captured in sex pheromone traps with different pheromone dispensers at Laki-hegy, 1979*

Date	Trap designs, pheromone concentrations and releasers			
	Pherocon 1C baited with 1 mg Codlemone in a Zoecon rubber cap	Reamol baited with 1 mg Codlemone in a Reanal rubber cap	Pherocon 1C baited with 1 mg Codlemone in a Swiss rubber tube	Sectar-like baited with 1 mg Codlemone in a Swiss rubber tube
a)				
16. 5.	9	2	—	—
23. 5.	35	1	—	—
30. 5.	13	6	—	—
7. 6.	13	2	—	—
12. 6.	1	0	—	—
20. 6.	3	0	—	—
26. 6.	4	0	—	—
3. 7.	0	1	—	—
10. 7.	7	8	—	—
Total	75	20	—	—
Mean of trap catches per week	8.3 <sup>a</sup>	2.2 <sup>a</sup>	—	—
b)				
17. 7.	8	1	3	2
24. 7.	3	0	4	1
31. 7.	0	2	5	1
7. 8.	2	2	7	0
14. 8.	0	0	0	1
21. 8.	6	0	1	2
28. 8.	1	0	0	0
4. 9.	1	0	0	0
12. 9.	1	0	0	0
18. 9.	1	0	0	0
25. 9.	0	0	0	0
3. 10.	0	0	0	0
9. 10.	0	0	0	0
16. 10.	0	0	0	0
Total	23	5	20	7
Mean of trap catches per week	1.64 <sup>a</sup>	0.36 <sup>a</sup>	1.42 <sup>a</sup>	0.5 <sup>a</sup>

(—) Means that the traps were not hung until after this date.

Data analysed by LSD test; means with different letters are significantly different at the 5% level.

## GENOTYPE-ENVIRONMENT INTERACTIONS IN THE MILK AND MEAT PRODUCTION OF CATTLE

Nowadays there is much talk of the importance of genotype-environment interactions and of the necessity of paying increased attention to them. The interaction between phenotype and environment means that the phenotypic variance of the population is induced not only by genetic and environmental effects but by the interactions between the two as well. If this interaction is of considerable extent it is likely to be manifested, so it is advisable to determine what conditions are necessary to induce a favourable interaction. The genotype-environment interaction is sometimes misinterpreted, too. Many people already speak of interaction when the phenotypic variances are different in different populations kept under identical conditions. This standpoint is disputable. If a genotype-environment interaction in the original sense of the word is to be spoken of, identical genotypes must be examined under different environmental conditions. If the difference in performance between animals with different genotypes in the same environment is considered, it is better to speak of different degrees of adaptation, but recently the responses obtained in such cases have also been counted among the genotype-environment interactions.

In the light of the various methods of interaction assessment four types of interaction can be differentiated:

- (a) one in which the genetic basis for the character is slight (low  $h^2$  value), while the different genotypes give hardly any response to the environmental effects (e.g. feed uptake);
- (b) low degree of genetic basis and high responsiveness to environmental effects;
- (c) high degree of genetic basis and poor response to environmental effects;
- (d) high degree of genetic basis and high responsiveness to environmental effects.

In this case the interaction is practically equal to zero.

Very few numerical data are available in the relevant literature on the genotype-environment interaction manifested in the milk production of cows, although many have studied the milk production of various breeds and crossing combinations, and the effects of various environmental factors. The assessment is complicated by the fact that the genetic variance of milk production is high while the covariance (genetic correlation) for milk production by the same genotype in different environment is low. Thus, the assessment will only give a significant result when a large stock of cows is examined, in which case the data processing is laborious. Researchers therefore usually content themselves with assessing the genetic variance of the stock and the genetic correlations and draw conclusions from these two on the genotype-environment interaction. Opinions formed in the international literature on the genotype-environment interaction for both milk production and cattle fattening are conflicting.

In milk and meat production, and in the characters related with them, the extent of interaction compared to the total variance is fairly small.

This summary has been drawn up on the basis of works by the following authors: RAVE (1974), LILJEDAHN *et al.* (1971), PIRCHNER (1972), AVERDUNK—ALPS (1974), RICHARDSON (1971), ROSS—CLAUSING (1974), ROSS—KRETZSCHMAR (1974), DOHY (1976), VÁGI—TÖRÖK (1975), KUNERT *et al.* (1974), DUNLOP (1962), VÁGI (1975), LANGHOLZ (1979), etc.

In 21 farms 1257 daughters of 27 bulls belonging to four different breeds or crossing combinations were examined for milk production, and 152 progenies of 9 bulls for meat production. The average size of the progeny groups was 46.3 for cows (extreme values: 21–67) and 16.9 for male animals. The culling proportion among the progeny was nearly the same (9–14%) in each case. The breeds and combinations included in the investigation were:

- Holstein-Friesian,
- Hungarian Fleckvieh × black spotted Holstein-Friesian  $F_1$
- Hungarian Fleckvieh × red spotted Holstein-Friesian  $F_1$ ,
- Hungarian Fleckvieh.

The bull progeny groups were kept in farms designated by the National Inspectorate of Animal Feeding and Breeding. The standard of feeding and keeping in these farms was higher than the average for Hungarian large-scale farms.

To determine the extent of interactions the total variance was broken down into variance components.

The following characteristics were examined:



**Table 1**  
*Significance of F-values of variance sources on the basis  
of variance analysis*

	Holstein- Friesian	Black spotted Holstein- Friesian × Hungarian Fleckvieh F <sub>1</sub>	Red spotted Holstein- Friesian × Hungarian Fleckvieh F <sub>1</sub>	Hungarian Fleckvieh
	1	2	3	4
<b>Milk yield</b>				
bulls	—	—	—	*
farm	—	—	—	—
interaction	—	—	*	**
<b>Butter-fat quantity</b>				
bulls	—	—	—	*
farm	*	*	—	**
interaction	*	***	—	—
<b>Persistence value</b>				
bulls	**	—	—	—
farm	—	—	—	—
interaction	***	***	—	—
<b>Lactation days, n</b>				
bulls	*	—	—	—
farm	***	—	—	—
interaction	—	—	—	—
<b>Age at first calving</b>				
bulls	*	—	—	—
farm	—	*	—	*
interaction	***	**	*	**
<b>Average daily body weight gain</b>				
bulls	*	*	—	**
farm	—	—	—	—
interaction	—	—	—	—
<b>Body weight on slaughtering</b>				
bulls	*	**	—	**
farm	**	*	—	**
interaction	*	*	—	*
<b>Bonned meat production</b>				
bulls	—	—	—	—
farm	—	—	—	—
interaction	—	—	—	—

\* P% = 5.0, \*\* P% = 1.0, \*\*\* P% = 0.1

- milk quantity,
- butter-fat quantity,
- persistence number,
- number of days in lactation,
- age at first calving,
- average daily increase in body weight,
- body weight on slaughtering,
- bonned meat production.

The table for the variance analysis of the bull progenies examined is not presented here since the SQ and MQ values do not give a great deal of information in themselves. Therefore, only data concerning the significance of F-values are included in Table 1. According to

**Table 2**  
*Breakdown of total variance*  
*(Variance components as percentages of total variance)*

	Holstein- Friesian cows	Black spotted Holstein- Friesian × Hungarian Fleckvieh F <sub>1</sub> cows	Red spotted Holstein- Friesian × Hungarian Fleckvieh F <sub>1</sub> cows	Hungarian Fleckvieh cows
	1	2	3	4
<b>Milk yield</b>				
$\sigma_{w_2}$ (error MQ)	—	—	80.84	68.22
$\sigma_{g_2 \times b_2}$ (farm × bull MQ)	—	—	1.76	10.82
$\sigma_{g_2}$ (farm MQ)	—	—	4.12	1.86
$\sigma_{b_2}$ (bull MQ)	—	—	13.28	19.10
<b>Butter-fat quantity</b>				
$\sigma_{w_2}$	88.18	84.49	—	—
$\sigma_{g_2 \times b_2}$	2.28	3.16	—	—
$\sigma_{g_2}$	0.79	1.76	—	—
$\sigma_{b_2}$	8.75	10.56	—	—
<b>Persistence value</b>				
$\sigma_{w_2}$	65.27	68.41	—	—
$\sigma_{g_2 \times b_2}$	17.73	7.28	—	—
$\sigma_{g_2}$	15.69	19.20	—	—
$\sigma_{b_2}$	1.31	5.11	—	—
<b>Lactation days, n</b>				
$\sigma_{w_2}$	—	—	—	—
$\sigma_{g_2 \times b_2}$	—	—	—	—
$\sigma_{g_2}$	—	—	—	—
$\sigma_{b_2}$	—	—	—	—
<b>Age at first calving</b>				
$\sigma_{w_2}$	76.20	70.26	78.31	73.32
$\sigma_{g_2 \times b_2}$	3.62	2.51	4.16	0.10
$\sigma_{g_2}$	8.96	17.56	10.63	7.30
$\sigma_{b_2}$	11.22	9.27	6.90	19.28

**Table 3**  
*Breakdown of total variance*  
*(Variance components as percentages of total variance)*

	Hungarian Fleckvieh	Holstein- Friesian × Hungarian Fleckvieh F <sub>1</sub>	Holstein- Friesian
	young fattening bulls		
Average daily body weight gain			
$\sigma_{x_1}$ (error MQ)	—	—	—
$\sigma_{g_1 \times b_1}$ (farm × bull MQ)	—	—	—
$\sigma_{g_1}$ (farm MQ)	—	—	—
$\sigma_{b_1}$	—	—	—
Body weight on slaughtering			
$\sigma_{x_1}$	74.90	74.42	79.15
$\sigma_{g_1 \times b_1}$	6.83	5.27	3.48
$\sigma_{g_1}$	11.36	7.86	12.24
$\sigma_{b_1}$	6.91	12.45	5.13
Boned meat production			
$\sigma_{x_1}$	—	—	—
$\sigma_{g_1 \times b_1}$	—	—	—
$\sigma_{g_1}$	—	—	—
$\sigma_{b_1}$	—	—	—

the data in the table, the F-value of the interaction is not higher than the F-value in the table for all the characters examined, nor for all breeds and combinations. Thus, the initial (zero) hypothesis, stating that no interaction exists between the present keeping and feeding conditions on large farms and the genotype, must be accepted in the following cases: with respect to milk yield in the Holstein-Friesian and Holstein-Friesian × Hungarian red spotted populations; with respect to butter-fat quantity and persistence in the red spotted Holstein-Friesian × Hungarian red spotted F<sub>1</sub> and the Hungarian red spotted populations; with respect to lactation period in all populations; and with respect to average daily weight gain and slaughter weight in all three populations examined.

**Table 4**  
*Ranking based on the production in groups of progenies from black spotted*

Farm	Number assigned to bulls							
	3540				3541			
	A	B	C	D	A	B	C	D
Milk yield	III.	I.	II.	IV;	III.	I.	II.	IV;
Butter-fat, kg	III.	II.	I.	IV;	III.	I.	II.	IV;
Age at first calving	I.	IV.	III.	II;	II.	IV.	III.	I;
Persistence	IV.	I.	II.	III;	III.	I.	II.	IV;
Lactation days, n	IV.	III.	I.	II;	II.	III.	IV.	I;



In Tables 2 and 3 the extent of interaction is shown by breaking down the variance to its components. According to the data in Table 2 the interaction in respect of milk quantity was 1.76% in the red spotted Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  progeny population and 10.82% in the Hungarian red spotted population. In the other two populations no interaction could be demonstrated for this character.

As regards the quantity of butter-fat the percentage of interaction was 2.28 in the Holstein-Friesian population and 3.16 in the Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  population. In the red spotted Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  and the Hungarian red spotted populations no interaction was found for the quantity of butter-fat.

The persistence index showed a noteworthy interaction between environment and genotype in two populations: 17.73% of the total variance in the Holstein-Friesian population and 7.28% in the Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  population was due to interaction.

With respect to the number of days in lactation no interaction could be demonstrated in any of the populations examined.

For the age at first calving, on the other hand, a non-significant but noteworthy interaction was manifested in all populations: 3.62% in the Holstein-Friesian stock; 2.91% in the black spotted Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  population; 6.16% in the red spotted Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  and 0.10% in the Hungarian red spotted population.

In Table 3 the interaction between the different genotypes and the Hungarian system of young bull fattening (intensive fattening using a dry feed mixture) are shown. As regards the average daily increase in body weight (live weight) an interaction of 5.27% was found in the Hungarian spotted population, 5.27% in the Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  and 3.48% in the Holstein-Friesian population (Table 3).

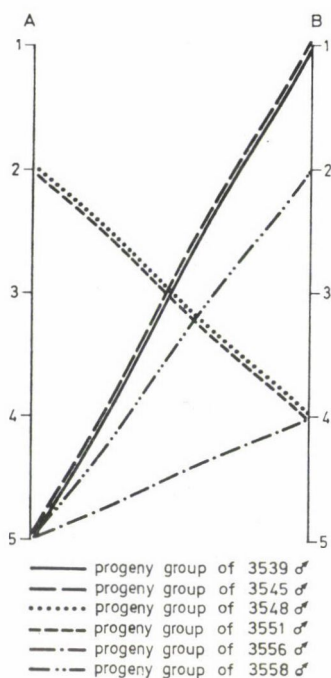
In Table 4 the bulls are ranked on the basis of the production of black spotted Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  progeny groups in various farms (Table 4). According to the data in the table the ranking of the bull progeny groups as regards the individual production parameters was fairly varied in the different farms. For example, it was not only with respect to the volume of milk yield, a parameter which is very dependent on environmental factors, but also for the quantity of butter-fat, that the order of the progeny groups was quite different in the farms. The data seem to suggest the existence of a considerable degree of interaction between the farms and the genotypes, which is not, however, verified by the analysis of variance.

Since the ranking of the bulls showed similar trends in the other three populations, the data will not be presented here.

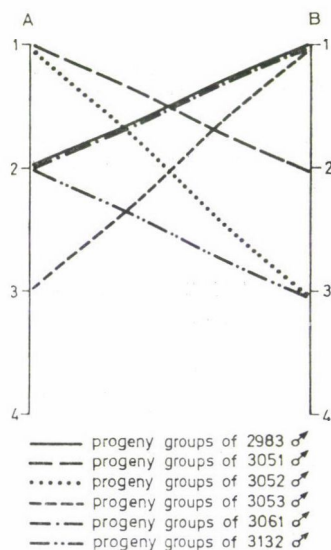
For the sake of illustration the ranking of certain genotypes, and the changes observed in it, are shown in Figs 1, 2 and 3 on the basis of production parameters from two farms. For example, in Fig. 1 the ranking of Holstein-Friesian bull progeny groups according to butter-fat production is quite different in the two farms (Fig. 1); two bulls placed second in Farm A on the basis of their progenies are placed fourth in Farm B. The changes in the ranking suggest that an interaction may exist. A similar trend is seen in the ranking of Hungarian red spotted bulls according to the lactation period of the progeny. There is a difference in the ranking between Farms A and B (Fig. 2).

#### *Holstein-Friesian $\times$ Hungarian Fleckvieh $F_1$ bulls ( $N = 282$ )*

3546				3554				3555			
A	B	C	D	A	B	C	D	A	B	C	D
II.	IV.	III.	I;	III.	IV.	I.	II;	III.	II.	I.	IV;
III.	IV.	II.	I;	IV.	III.	I.	II;	II.	II.	I.	IV;
I.	II.	IV.	III;	I.	IV.	III.	II;	II.	IV.	I.	III;
III.	I.	IV.	II;	III.	I.	II.	IV;	II.	I.	III.	IV;
III.	IV.	II.	I;	III.	II.	I.	IV;	II.	IV.	I.	III;



**Fig. 1.** Order of progeny groups of Holstein-Friesian bulls in Farms A and B according to butter-fat production



**Fig. 2.** Order of progeny groups of Hungarian Fleckvieh bulls in Farm A and B according to the number of days in lactation

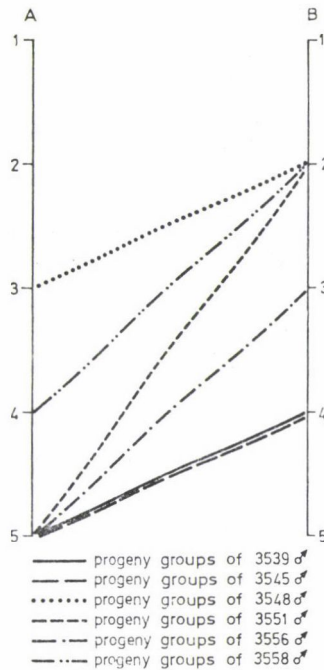


Fig. 3. Order of progeny groups of Holstein-Friesian bulls in Farms A and B according to the persistence index

According to Fig. 3 the classification of bulls on the basis of the production results achieved by the progenies shows only minor changes. In Farm B all the bulls occupy more favourable places than in Farm A (Fig. 3).

The interaction represented in Figs 1, 2 and 3 is not always a true picture of the actual situation. For example, the data in Figs 1 and 2 suggest the existence of interaction, whereas the analysis of variance shows no interaction between these data. On the basis of Fig. 3 there would appear to be no interaction, yet according to the variance analysis it is here that the interaction between genotypes and farms is the closest.

Prior to evaluating the data, it should be noted that, owing to the mathematically small number of specimens included in the study, the assessment is not efficient enough (although from a biological point of view this population was sufficiently large to draw conclusions from). If the arrangement had been more uniform, i.e. the difference between the groups (populations) in the number of observations had been smaller, the size of the interactions might have been modified. At the same time, it must not be forgotten that a large proportion of the available literature reports on results obtained with much smaller experimental stocks.

Cattle populations belonging to four different breeds or crossing combinations were studied in order to determine the extent of interaction which is to be expected between the keeping and feeding technologies on Hungarian large-scale farms and the different genotypes.

On the basis of the experimental results the first question to be settled is whether any substantial degree of interaction is to be expected under large-scale keeping and feeding conditions. According to the experimental data, as regards the volume of milk yield no interaction existed in the Holstein-Friesian or the Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  populations and a very slight interaction in the red spotted Holstein-Friesian  $\times$  Hungarian red spotted  $F_1$  generation; only the progeny groups of Hungarian red spotted bulls showed an interaction of around 11%. In the case of the other qualities examined there was either no interaction or it represented only a few percent. A noteworthy interaction was only found for the persistence index of the Holstein-Friesian breed.



The situation is similar for the interactions manifested in meat production. It should be noted, however, that these results refer to the dry feed mixture technology of fattening.

It is felt that selection or other breeding work aimed at utilizing the interaction between genotype and environment is only justified when the proportion of interaction is at least 15–20% of the total demonstrable phenotypic variance. Considering the current efforts to standardize the environmental conditions such a high extent of interaction is unlikely to develop, and its induction is not justified without preliminary economic analyses.

The second question to be answered on the basis of the experimental data is whether changes in the rankings of the bulls influence the reliability of sire evaluation to such an extent that the interaction should be incorporated in the evaluation. According to the data the genotype-environment interaction in the breeds and combinations kept under large-scale farm conditions in Hungary is of minor importance; there is therefore no justification for including it in the sire evaluation either now or in the near future. In support of this statement it should be noted that the present tendency is to consolidate the characters of all breeds reared in Hungary. The genetic variance of the bulls will thus decrease; consequently in an increasingly uniform and optimized environment the extent of interaction manifest in these characters will also presumably decrease in the future.

Finally, these investigations also give some information on whether the genotypes examined require different environmental conditions to achieve their production potentials.

In the light of the fact that for several characters no interaction was manifested, and that even where interaction could be demonstrated its effect was very slight, it seems that the tendency to create optimum environmental conditions meets the requirements of all the genotypes examined. This is all the more the case, since these populations are genetically ahead of the present environmental conditions in Hungary.

From the experimental data it can be concluded that the reliability of sire evaluation will not decrease if interactions are ignored. Optimum environmental conditions do not necessarily increase phenotypic variance, so it is worth creating such conditions in the interests of sire evaluation. Breeding aimed at utilizing the interaction between genotype and environment ("systematic induction of desirable interactions") is only justified when the variance due to interaction makes up at least 15–20% of the total variance.

\*

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### References

- AVERDUNK, G.—ALPS, H. (1974): Die Interaktion zwischen Herdenniveau und Vater bei der Milchleistung des Fleckviehs in Bayern. (Manuscript.)
- DOHY, J. (1976): Adatok és gondolatok a genotípus és a környezet kölcsönhatásának problémaköréhez (Contribution to and reflections on the question of genotype-environment interaction). Magyar Állatorvosok Lapja, **10**, 645–649.
- DUNLOP, A. A. (1962): Interaction between heredity and environment in the Aust. Merino. Austr. J. Agric. Res., **13**, 503–531.
- KUNERT, G.—KUNERT, H.—SCHWARK, H. J. (1974): Leistungsteigerung und Erbllichkeit von Milchleistungseigenschaften beim Rind. Inst. Landwirt. Inf. Dok., Berlin, **12**, 12.
- LANGHOLZ, H. J. (1979): The impact of genotype-environment interaction on breeding and production planning in cattle. EAAP, Harrogate.
- LILJEDAHN, L. E.—HENNINGSSON, T.—LUNDKVIST, G. (1971): Studies on monozygous cattle twins. Swedish J. Agric. Res. Stockholm. 1971. 1. 3. sz. 191–201
- PIRCHNER, F. (1972): Interaktion zwischen Genotyp und Mastverfahren bei Rindern. Tierzüchter, **24/24**, 712–714.
- RAVE, G. (1974): Genotyp-Umwelt-Interaktion im Rahmen der Fleischleistung beim Rind. Tierzüchter, **26/4**, 150–152.
- ROSS, K.—CLAUSING, P. (1974): Korrekturfunktionen zur Beseitigung von Effekten unterschiedlicher genotypischer Konstruktionen für die Verbesserung der Zuchtwertschätzung beim Rind. Karl Marx Univ., Berlin, 266–272.

- ROSS, K.—KRETZSCHMAR, B. (1974): Genotyp-Umwelt-Interaktionen bei Milchleistungsmerkmalen und deren Auswirkungen auf die Züchterwertschätzung von Bullen. Karl Marx Univ., Leipzig, 255–261.
- VÁCI, J.—TÖRÖK, I. (1975): A genotípus  $\times$  takarmányozási időszak változásai kölcsönhatásának vizsgálata (Interaction between genotype and feeding period). Agrártud. Egyetem Közleményei, Gödöllő.
- VÁCI, J. (1975): Szarvasmarha hizodalmasság vizsgálatok a genotípus–környezet kölcsönhatások figyelembe vételére alkalmas kortársas ivadékvizsgálati módszerrel (Propensity to fattening in cattle as studied by contemporary progeny tests which take genotype–environment interactions into consideration). Thesis, Gödöllő.





## LECTIONES

### CHANGES IN THE PRODUCTION OF MAIZE HYBRIDS DUE TO MUTANT PARENT LINES\*

Improving the quantity and quality of yields is a basic and permanent aim in agriculture. This aim can only be reached by developing the biological background of plant breeding and modernizing the various agrotechnical procedures.

In comparison with the amount of plant production theoretically possible by photosynthesis, only a small proportion is realized in practice. It is the task of plant breeders to increase the intensity and the production of photosynthesis by varieties of modern genetical composition.

In plants it is the size of the leaf area that basically determines the synthesis of organic matter. Experiments were conducted to see how parent lines, developed by irradiation-induced mutation, modify the size and the productivity of the assimilation area in hybrids. The mutant lines that we developed were tested for varieties that can be used for producing hybrids of a genetic structure that enables them to utilize solar energy more efficiently, and also show a positive heterosis effect in yields. Several theories have been put forward as to the genetical mechanism of heterosis in maize (BÁLINT 1967, SVÁB 1971, LE ROY 1972).

A number of studies explain the above-mentioned phenomenon by changes occurring in the basic metabolisms.

An examination of the phylo-physiological differences can bring out the superiority of hybrids over their parent lines in various ways.

Of the factors affecting photosynthesis, hybrids are superior to parent lines in the size of the leaf area (DOBROVSKAJA 1962) (in FJODOROV 1968). A similar conclusion can be drawn from an examination of the dry weight of leaves at various stages of development (FJODOROV 1968).

If the biological superiority of maize hybrids is to be ensured, a larger assimilation area should be accompanied by a greater efficiency of photosynthesis (FJODOROV 1968).

In order to find the optimum of organic matter production, the relation between assimilation and dissimilation should be studied. The relation between assimilation and dissimilation in various maize hybrids and their parent lines was studied by SERBANESCU (1966). He found that photosynthesis is more intensive in two-line hybrids than in their parent lines. Dissimilation losses are smaller in hybrids than the average of their parent lines. GÖRING's (1963) findings are similar, too. He found that oxygen consumption (in gram per dry matter) was greater in the parent lines than in the hybrids. GÁSPÁR (1963) found differences between parent lines and hybrids even in root respiration.

The intensity of photosynthesis does not only vary from one species to the next; varieties, too, can differ significantly in this respect. Differences in the foundation seed stocks should result in higher yields among the hybrids.

The productivity of photosynthesis can be improved by selecting and crossing varieties of good characteristics. The production of organic matter and the efficiency of assimilation are determined by a number of morphological and physiological factors. Of these the size of the leaf area is of great importance. Increasing dry matter yield and effective yield requires that maize hybrids have erect leaves (LOOMIS and WILLIAMS 1972), and that their leaf area indices also be increased (DUNCAN 1975).

\* Lecture held at the meeting of the ESNA (European Society of Nuclear Methods in Agriculture) in Brno, CSSR, from 6th September to 11th September 1982.

The aim of maize breeders is to produce hybrids that are ideal from the standpoint of effective yield production. There is no direct correlation found between productivity and the net assimilation rate (NAR), because assimilates are distributed unevenly among the various plant parts (LUPTON 1962). The superiority of hybrids is revealed in the more favourable proportions of the generative organs, too (GYÖRFFY 1962). Dry matter yield per unit leaf area is higher in hybrids than in open pollinated varieties (BELLINI and FUSI 1966).

Several studies have been published on factors affecting the size of the leaf area. WATSON (1956), BLACKMAN *et al.* (1955), BLACK (1955) report on the relation found between the assimilation rate and the leaf area index. The relation between plant density, leaf area and productivity was studied by BEZRUKOVA (1965), MOZSAEV (1966), BAJAI (1959) and MAUL *et al.* (1964). EIK, K. *et al.* (1966) published his findings on the correlation between the leaf area index (LAI) and prospective yield.

MENYHÉRT—ÁNGYÁN—RADICS (1980) and TOUNI (1968) studied the various factors determining the size of the leaf area in detail.

### Materials and methods

Our experiment was conducted at the Debrecen-Kismacs part of the Experimental Station of the Agricultural University of Debrecen. The parent lines and hybrids to be studied were the following:

Mothers ( $P_1$ ): MQ<sub>2</sub>-1, DPM 2-59, SC 2390

Fathers ( $P_2$ ): M-93, M-206, M-197

Hybrids ( $F_1$ ): SC-2390, MQ<sub>2</sub>-1  $\times$  M-93, DPM 2-59  $\times$  M-93

SC-2390  $\times$  M-93, SC-2390  $\times$  M-206, SC-2390  $\times$  M-197

Pi 3764 MSC, Beke 370 TC, Mv SC 434, Pi 3709 MSC

Hybrid SC-2390, which is mentioned both among the hybrids and among the parents, had an (MQ<sub>2</sub>-1) Opaque mutant as its maternal line. Its paternal line (DPM 2-59) was selected from an inbred line that had been irradiated during the vegetation period by a dose of 7 Gy (of a <sup>60</sup>Co radiation source) in the gammafield of the Agricultural University of Gödöllő. "M" marks paternal lines ( $P_2$ ) selected from mutant populations, that were produced in 1958 from local strains by treating their pollens with a dose of 15 Gy (of <sup>60</sup>Co).

Leaf area was determined by Montgomery's method, as this was found to be the most suitable one to conditions in Hungary (PINTÉR 1979). The length and width of leaves were measured in 4 replications on five plants each time.

All the hybrids and paternal lines underwent production-biological examinations at the silage stage in 4 replications, using 5 plants at each replication.

Mathematical-statistical data processing was done by variance-analysis, factor-analysis and cluster-analysis.

Variance-analysis was used to see if there were any significant differences between the hybrids and their parent lines in a number of variables. Factor analysis was done to determine the variables that most effect NAR. Hybrids and their parent lines were classed, and the classes were illustrated by a treegraph (or dendrogram) in an orthogonal co-ordinate system. Our data were cluster-analysed following standardization.

Variance-analysis was done by the method developed by VINCZE (1975) and SVÁB (1981), while cluster-analysis was done following the method developed by FÜSTÖS—ME-SZÉNA—MOSOLYGÓ (1977), PODANI (1980) and RÉVÉSZ—FRITZ (1974).

### Results of the experiments

The results of experiments, started in 1981 with mutant parent lines of various geno-types as well as two- and three-line hybrids, are as follows:

#### *Leaf area and leaf area index*

Leaf areas of various mutant parent lines and their hybrids were measured three times (on May 15th, June 12th and July 9th). The results are shown in Table 1.

The data of Table 1 show that mutant lines have extremely large leaf areas and that this trait is inherited by their progeny (hybrids 4, 5, 10 and 11).

When two-line hybrids (4, 5 and 6) were compared with standard two-line hybrids (12, 13 and 15), it was found that (except for one case) hybrids 4, 5 and 6 had significantly larger leaf areas on May 15th. On June 15th hybrids 4 and 5 significantly exceeded leaf areas



**Table 1**  
*Leaf areas and leaf area indices of the parents and their hybrids*  
(Debrecen, 1981)

Parents and their hybrids	Average size of leaf area, cm <sup>2</sup> /plant			Leaf area index (LAI), m <sup>2</sup> /m <sup>2</sup> soil
	May 15th	June 12th	July 9th	
1. MQ <sub>2</sub> -1	197.5	2377.4	5727.1	4.09
2. DPM 2-59	170.3	2070.5	5076.1	3.63
3. M-93	269.5	2856.3	6010.3	4.29
4. MQ <sub>2</sub> -1 × M-93 (SC)	348.3	3875.4	6407.2	4.82
5. DPM 2-59 × M-93 (SC)	316.7	3610.5	6295.8	4.57
6. SC 2390	247.6	3015.8	5960.4	4.26
7. M-206	240.3	2605.3	5926.3	4.23
8. M-197	221.1	2419.5	5805.4	4.15
9. SC 2390 × M-93 (TC)	380.4	4125.5	7225.7	5.16
10. SC 2390 × M-206 (TC)	350.6	3957.5	6885.4	4.92
11. SC 2390 × M-197 (TC)	335.9	3831.6	6665.2	4.68
12. Pi 3764 MSC	265.7	3348.6	6122.8	4.37
13. BEKE 370 (TC)	254.9	3354.7	5963.2	4.26
14. MVSC 434	170.2	3234.2	5883.6	4.20
15. Pi 3709 MSC	172.8	2582.3	5985.6	4.49
SD <sub>5%</sub>	14.3	185.1	373.3	0.26

of the three standards (12, 13 and 15). On July 9th no significant difference was found between the leaf area of standard Pi 3764 MSC and that of the two-line hybrids. When compared with standard MVSC 434, only hybrids 4 and 5, while compared with standard Pi 3709 MSC only hybrid 4, showed significant difference in leaf area. Leaf areas of three-line hybrids significantly exceeded that of standard BEKE 370 (TC) at all three times of measurement.

Of all two-line hybrids, leaf area index (LAI) was found highest in hybrid 4 compared with any of the standards. Hybrid 5 had a higher value of LAI only in comparison with standard MVSC 434. The value of LAI was significantly higher in all three-line hybrids than in standard BEKE 370 (TC). Table 2 illustrates heterosis effect in leaf area and presents averages of leaf area, and their differences compared with parent lines.

The data of Table 2 show significant heterosis effect in all hybrids.

#### *Productivity in parent lines and in hybrids*

Table 3 shows net assimilation rate (NAR), productivity and ratio of effective yield, of hybrids harvested at silage stage.

The data of the table show that two-line — and especially three-line — hybrids have significantly higher net assimilation rates (NAR) than the parent lines or the standard hybrids. Total yield per 1 m<sup>2</sup> is especially high in three-line hybrids, whose paternal lines were mutants. Three-line hybrids SC 2390 × M-93 and SC 2390 × M-206 excel in grain yield per unit leaf area. The proportion of effective yield is highest in parent lines MQ<sub>2</sub>-1 and DPM 2-59, in hybrid SC 2390 and standard hybrids MVSC 434 and Pi 3719 MSC.

#### *A mathematical-statistical evaluation of the results*

Variance analysis showed that the difference in the examined characters between the hybrids and their parent lines is significant at a level of 0.1% (Tables 1 and 3, Fig. 1).

The following characters of the plants were examined by factor analysis: leaf area on July 9th, leaf area index, net assimilation rate (NAR), productivity, total yield and grain yield per 1 m<sup>2</sup> leaf area. The correlation coefficients in Table 4 show that the examined characters are closely related.



**Table 2**

*Heterosis in the leaf area of SC and TC hybrids originating from mutant lines  
(Debrecen, 1981)*

Hybrids	Average value of leaf area								
	May 15th			June 12th			July 9th		
	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$
	cm <sup>2</sup> /plant		$\bar{P}$	cm <sup>2</sup> /plant		$\bar{P}$	cm <sup>2</sup> /plant		$\bar{P}$
MQ <sub>2</sub> -1 × M-93 (SC)	233.5	114.8	0.492	2616.9	1258.5	0.481	5868.7	538.5	0.092
DPM 2-59 × M-93 (SC)	219.9	96.8	0.440	2463.4	1147.1	0.466	5543.3	752.5	0.131
SC 2390	183.9	63.7	0.346	2224.0	791.8	0.356	5401.7	558.7	0.103
SC 2390 × M-93 (TC)	258.6	121.8	0.471	2936.1	1189.4	0.405	5985.4	1240.3	0.207
SC 2390 × M-206 (TC)	244.0	106.6	0.437	2810.6	1146.9	0.408	5943.4	942.0	0.159
SC 2390 × M-197 (TC)	234.4	101.5	0.433	2717.7	1113.9	0.410	5882.9	682.3	0.116

**Table 3**

*NAR productivity and rate of effective yield in parent lines of various genotypes  
and in their hybrids (at silage stage)*

Parent lines and hybrids	Net assimila- tion rate (NAR), g/plant	Productivity 1 m <sup>2</sup> leaf area in grams		Rate of effective yield, %
		total yield	grain yield	
MQ <sub>2</sub> -1	214.0	373.7	136.0	36.4
DPM 2-59	203.6	401.1	144.8	36.1
M-93	287.8	478.8	126.9	26.5
MQ <sub>2</sub> -1 × M-93 (SC)	316.3	493.7	151.6	30.7
DPM 2-59 × M-93 (SC)	312.5	496.4	153.9	31.0
SC 2390	270.1	453.2	165.4	36.5
M-206	293.5	495.3	142.2	28.7
M-197	195.8	509.5	143.7	28.2
SC 2390 × M-93 (TC)	419.0	579.9	190.2	32.8
SC 2390 × M-206 (TC)	396.5	575.9	186.6	32.4
SC 2390 × M-197 (TC)	369.4	562.7	180.6	32.1
Pi 3764 MSC	305.2	498.5	164.5	33.0
BEKE 370 TC	309.3	518.7	175.3	33.8
MVSC 434	286.5	486.9	183.6	37.7
Pi 3719 MSC	283.2	473.1	181.2	38.3
SD5%	13.9	21.4	8.0	1.2

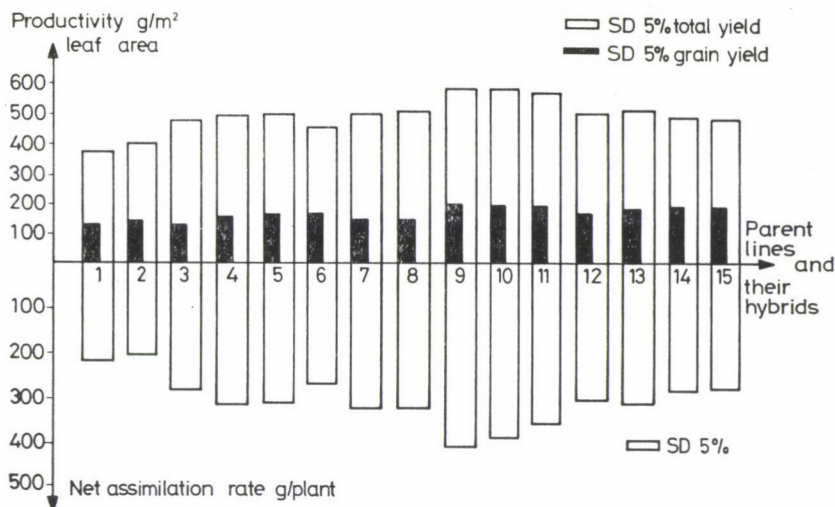


Fig. 1. NAR of silage, Productivity and rate of effective yield in parent lines of various genotypes and in their hybrids

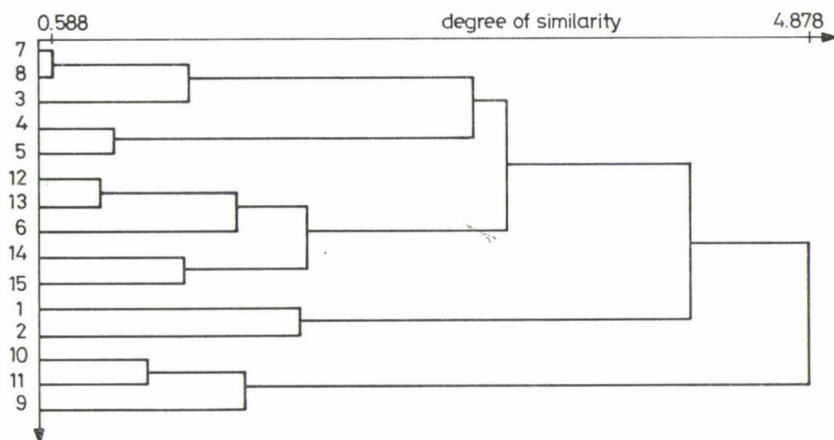


Fig. 2. A classification of parent lines and hybrids by cluster analysis of 8 variables

The variables had two common factors, which accounted for 91.76% of the variance. After appropriate transformation of the factor matrix, the characters that most affect the net assimilation rate could be determined. Our findings show that these are the size of the leaf area and the leaf area index.

Cluster analysing the afore-mentioned 8 characters, we could classify hybrids and their parent lines in the following way:

- Class 1. 3. M-93  
7. M-206  
9. M-197
- Class 2. 4. MQ<sub>2</sub>-1 × M-93  
5. DPM 2-59 × M-93

- Class 3. 6. SC 2390  
 12. Pi 3764 MSC  
 13. BEKE 370 TC  
 14. MVSC 434  
 15. Pi 3709 MSC
- Class 4. 1. MQ<sub>2</sub>-1  
 2. DPM 2-59
- Class 5. 9. SC 2390 × M-93  
 10. SC 2390 × M-206  
 11. SC 2390 × M-197

Table 4

*Values of the correlation coefficients and the rotated factor matrix*

Variables	1	2	3	4	5
1. Leaf area (July 9th)	1.00000	0.99400	0.86173	0.73275	0.53934
2. Leaf area index	0.99400	1.00000	0.87086	0.74724	0.54292
3. Net assimilation rate	0.86173	0.87086	1.00000	0.91880	0.61138
4. Productivity per 1 m <sup>2</sup> leaf area total yield	0.73275	0.74724	0.91880	1.00000	0.58878
5. Productivity per 1 m <sup>2</sup> leaf area grain yield	0.53934	0.54292	0.61138	0.58878	1.00000
Rotated factor matrix					
Factor No. 1	0.94876	0.95459	0.96387	0.89391	0.66905
Factor No. 2	-0.19054	-0.18607	0.00000	0.08749	0.72282

### Summary

Assimilation leaf area and productivity of hybrids, originating from mutant lines of various genotypes, and of their parent lines, were studied in small-plot comparative experiments by researchers of the Plant Production and Ecology Institute of the Agricultural University of Debrecen. The results were evaluated by mathematical-statistical methods. Our findings can be summed up as follows:

- The mutant lines that were used as paternal lines in crossings had large leaf areas, which is advantageous to photosynthesis.
- The assimilation leaf areas of hybrids originating from crosses of certain mutant lines showed significant heterosis effect, when examined at various times.
- The value of leaf area index (LAI) was higher in SC and TC hybrids than in their mutant parent lines.
- Total yield and grain yield per 1 m<sup>2</sup> leaf are increased significantly in three-line hybrids originating from mutant lines, when harvested at silage stage.
- The proportion of effective yield was good in some parent lines and their hybrids. It decreased, however, in three-line mutants originating from mutant lines.
- The value of the net assimilation rate (NAR), as well as the balance of assimilation and dissimilation, improved in two- and three-line hybrids originating from mutant lines.
- The results of our experiments lead us to believe that mutants are best used as paternal lines in crosses producing three-line hybrids.
- Variance analysis of the data showed significant differences among all the characteristics of the hybrids and their parent lines at a level of 0.1%. Factor analysis showed that the net assimilation rate is significantly affected by the size of the leaf area index. Cluster analysis enabled us to set up 5 classes for the hybrids and their parent lines on the basis of the studied 8 variables.

\*

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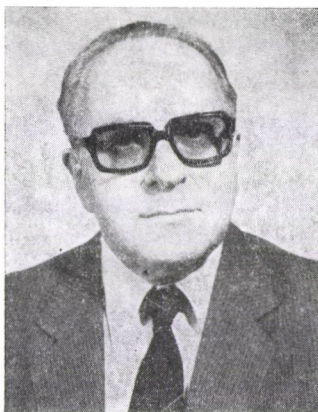


## References

- BAJAI, J. (1959): Összefüggés a kukorica levélfelülete és a tenyészterület különféle alakja között. *Növénytermelés* S/3. 217–221.
- BÁLINT, A. (1967): Heterózis és mutáció a kukoricában. Akadémiai Kiadó, Budapest.
- BELLINI, P.—FUSI, G. (1961): Rese individuali ed unitarie dei diversi organi epigei in *Zea mays* L. *Elette*. VII: 1: 29–40.
- BEZRUKOVA, V. P. (1965): A növényesűrűség hatása a levélfelületre és a szemtermésre. (Vlijanis gusztotij poszeva na razvitie lisztovoj poverhnoszi is uroszaj zerna.) *Kukuruza*, Moszkva. 1965. (10), 12, 26–67.
- BLACK, J. N. (1955): The interaction of light and temperature in determining the growth rate of subterranean clover (*Trifolium subterraneum* L.). *Aust. Journ. Biol. Sci.*, **8**, 333–343.
- BLACKMAN, G. E. *et al.* (1955): Physiological and ecological studies in the analysis of plant environment. *Ann. Bot.*, New Series, **19**, 527–548.
- DUNCAN, W. G. (1975): Maize. In: EVANS, T.: *Crop physiology*. Cambridge Univ. Press, London—New York. 23–50.
- EIK, K. *et al.* (1966): Leaf area in relation to yield of corn grain. *Agron. Jour. Madison*, 1966. **58**, (1): 16–18.
- FJODOROV, P. Sz. (1968): Biohimicseszkie i fiziologicseszkie osnovü geterozisza kukuruzii. *Uzg. Kirgizisztan, Frunze*.
- FÜSTÖS, L.—MESZÉNA, GY.—S. MOSOLYÓ, N. (1977): Cluster analízis. *Sigma* **10**, 111–148.
- GÁSPÁR, L. (1963): Contribution to the biology of hybrid vigour. In: *Symposium on Maize Breeding and Production*. Martonvásár, 59–66.
- GÖRING, H. (1963): Respiratory particularities of germinating seeds of inbred maize stocks and hybrids. In: *Symposium on Maize Breeding and Production*. Martonvásár. 67–70.
- GYÖRFFY, B.—I'SÓ, I.—BÖLÖNI, I. (1965): *Kukoricatermesztés*. Mezőgazdasági Kiadó, Budapest.
- LE ROY, H. L. (1972): Heterosis in Population. *Zeitschrift für Pflanzenzüchtung*. 67, 1, 65–78.
- LOOMIS, R. S.—WILLIAMS, W. A. (1972): Plant conformation and yield. In: *Induced mutation and plant improvement*. IAEA, Vienna, 13–25.
- LUPTON, F. G. G. (1966): Translocation of photosynthetic assimilates in wheat. *Ann. Appl. Biol.* **57**, 337–364.
- MAUL, F.—PITYINGER, O. (1964): Az állománysűrűség hatása a kukorica asszimiláló felületre és csőtermésére Mátra—Bükk aljai csernozjom barna erdőtalajon. *Növénytermelés* **13** (2), 131–138.
- MENYHÉRT, Z.—ÁNGYÁN, J.—RADICS, L. (1980): A levélfelület-index (LAI), a fényviszonyok és a termés kapcsolata eltérő vetésidőjű és tenyészterületű kukorica állományokban. *Növénytermelés*. Tom. 29, No. 4.
- MOZSAEV, N. I. (1966): A levélfelület területe és a silókukoricatermés. (Ploscsad lisztovoj Poverhnoszi i uroszaj kukuruzii na szilosz.) *Kukuruza*, Moszkva, **8**, 13–14.
- PINTÉR, L. (1979): A kukorica (*Zea mays* L.) hibridek levélfelületének gyors meghatározási lehetőségei hazai biológiai viszonyok között. *Növénytermelés*, **28**, 397–401.
- PODANI, J. (1980): SYN-TAX számítógépes programcsomag ökológiai, cönológiai és taxonómiai osztályozások végrehajtására. Az ELTE Növényrendszertani és Ökológiai Tanszékének Kiadványa 158 p.
- RÉVÉSZ, P.—FRITZ, J. (1974): Az alakfelismerés statisztikus módszerei. Az MTA Matematikai Kutató Intézetének jegyzete, Budapest.
- SERBANESCU, E. (1966): Cercetari fiziologice la plante hibride si la formele lor parantale. *Studii si cercetari de biologie*. **18/5**, 461–469.
- SVÁB, J. (1971): A populációgenetika alapjai. Mezőgazdasági Kiadó, Budapest.
- SVÁB, J. (1981): Biometria módszerek a kutatásban. Mezőgazdasági Kiadó, Budapest 557 p.
- TOUNI, A. H. (1968): Különböző tényezők hatása a kukorica levélfelületének nagyságára és szemtermésére. *Növénytermelés*, **2**, 139–149.
- VINCZE, I. (1975): Matematikai statisztika. Az ELTE Természettudományi Karának jegyzete, Budapest 367 p.
- WATSON, D. J. (1956): Leaf growth in relation to crop yield. In: MILTHORPE, F. L. (ed.): *The growth of leaves*, pp. 177–191. London, England, Butterworths publications, Ltd.



## AS I SEE IT...



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### THE FUTURE OF HYBRID MAIZE BREEDING IN HUNGARY

Before discussing the future of Hungarian maize breeding, I feel it is necessary to make a short retrospect into its recent past and to survey its present. It will not be worth outlining the future until having weighed up the possibilities of maize breeding in the temperate zone.

The importance of maize in Hungary can be emphasized by several data. Maize is the most important field crop in Hungary. In 1976-1980 it was grown on 32.4% of the arable land for grain and silage and it gave 50.5% of the whole corn and cereals production. Including exported seed, the gross production value was 24.2 billion forints. This value was 16.8% higher than the value of the yield for cereals. Maize gives approx. 70% of the Hungarian feed grain requirement. In the protein balance it has a proportion of 40%, because of its great volume. (It is interesting to note that the gross production value of the maize yield calculated in the above manner—on the average of 1981 and 1982, two very favourable years, was more than 27.5 billion forints.) The effect of maize breeding can hardly be overestimated even if we only regard the increase in average yield due to the yearly 1.0-1.3% genetic contribution as a standard. This means approx. 300 million forints annually.

### Retrospect

The news of the pioneer work in hybrid maize breeding in the US as the result of inbreeding reached Hungary by the 1930s. Rudolf Fleischmann, working in Kompolt (Heves county) as a state researcher, had produced his own inbred lines by the end of the 1930s. At the same time, Endre Pap, as a private farmer and plant breeder in Mindszentpuszta (Fejér county), evolved inbred lines from his own varieties in order to create the necessary conditions for the utilization of the heterosis effect. The work of Rudolf Fleischmann was continued by Ferenc Szüllő in Bánkút (Békés county), who passed on the inbreds developed from the Fleischmann varieties to a research group in Keszthely at the beginning of the 1960s, where László Berzsenyi-Janosits directed research at the Agricultural University. In addition, inbred lines had been evolved by Ferenc Beke in Fertőd, Lajos Daniel in Budapest, Miklós Horn in Lovászpátona—just to mention the most important of those who did not continue commercial hybrid maize breeding after the sixties. Most of these inbred lines were taken to Keszthely after the reorganization of agricultural research at the beginning of the 1960s, while some of them have been used in practical breeding in Martonvásár.

For several years contacts with US maize breeders was interrupted due to the political changes which occurred at the end of the 1940s. During that period French and Yugoslavian



breeders were not only able to learn about heterosis breeding from American researchers who visited these countries, but also had the possibility of spending a year or so in the U.S.A. There they could study on the spot the most important breeding methods and the well-tried technologies for maintenance breeding, parent seed multiplication and commercial seed production. It was not easy for Hungarian research workers, only very few of whom had the opportunity to visit the U.S.A. in the 1950s and 1960s, to overcome this handicap. Regular visits to American universities and breeding firms have only been possible since the 1970s. In spite of this it is worth noting that Hungarian maize breeding played a pioneer role in many areas in the 1950s and 1960s in Europe. The reason was that the work of the two outstanding researchers (R. Fleischmann and E. Pap) was partly in advance of that of most European breeders, and was partly carried out at the same time. (Internationally outstanding results were attained by Hadjinov in Krasnodar, by Tavcar in Zagreb and by Cauderon in Versailles.) The other reason, in my opinion, is that many of the American public lines became available to Hungarian breeders by the end of the 1940s and in the 1950s.

In the 1950s Endre Pap and László Berzsenyi-Janossits were the two leading maize breeders in Hungary. While Endre Pap, who had been working at the Agricultural Research Institute of the Hungarian Academy of Sciences in Martonvásár since 1950, dealt exclusively with inbred hybrid maize breeding, László Berzsenyi-Janossits achieved longstanding results in Mosonmagyaróvár chiefly on the possibilities latent in variety heterosis. Many variety hybrids such as Óvári 1, Óvári 4 and Óvári 5, were linked with his name, and these were widely grown at that time.

The series of successes in inbred hybrid maize breeding, which became famous all over Europe, was begun in 1953 by the registration of the double-cross hybrid Mv 5. What were the reasons for this successful overture? The most important reason perhaps, was that the most promising breeding material was in the hands of an exceptionally intelligent and creative breeder. The Mindszentpusztai yellow dent variety had a very wide genetic basis. Its strength lay in its yielding ability and relatively early maturity. Due to this fact Endre Pap obtained outstanding inbred lines, such as No. 156, 0118b, 014, etc., which showed a good heterosis effect even when crossed with each other. On the basis of later results there is every justification for assuming that the Mindszentpusztai yellow dent variety, the breeding of which was started in Mindszentpuszta from a yellow dent maize population from Bács county by Áron Pap and Endre Pap in 1917, could only have been distantly related to the US varieties Wilson Farm Reid, Hayes Golden, Golden Glow, etc., from which the inbred lines WF9, N6, W23 (C5) were bred. This fact is proved by the good yielding ability of the hybrid Mv 5, which originated from the American inbred line W23 (called C5) as well as from Mindszentpusztai lines. The DC hybrid Mv 1 registered 3 years later showed even higher yielding ability. Among its parents we find the American inbred lines WF9 and M14 as well as C5. The Hungarian "blood" is represented by the inbred line 014.

The creativity of the Martonvásár breeders led by István Kovács, who took over from Endre Pap, is demonstrated by the fact that they substituted the inbred line M14, which was susceptible to *Fusarium*, by the more up-to-date inbred line N6. The use of this line, together with the more and more intensive use of line No. 156 resulted in the official registration of the hybrids Mv 59 (in 1962) and Mv 602 DC (in 1967). Though these hybrids suited the maize production of the 1960s very well, it was nevertheless the single-cross hybrids Mv SC 530, Mv SC 570 and Mv SC 620 registered in 1968 that represented the most significant progress. It may be of interest to mention that the Hungarian inbred line No. 156, the female parent of Mv SC 530, again proved to be the most successful of the four which made up these hybrids, preceding the three inbred lines from the U.S.A.

Another striking success of the inbred hybrid maize breeding in Martonvásár was the hybrid Mv SC 580 registered in 1972. This was the most characteristic hybrid of the FAO 500-599 maturity group for 11 years. In this case the line No. 156 resulted in an excellent hybrid with the inbred line B14, which was an outstanding representative of the inbred lines following WF9, M14, etc. (Just by way of introduction it may be added that the numerous sisterlines of B14 still play a decisive role in maize production in the temperate zone, demonstrating particularly the general success of the back-cross and recurrent selection methods.) It is very probable that the source material of these two outstanding inbred lines became separated from each other many centuries ago and passed through significant introgressions. Moreover, it can be assumed that the different selection effect of the environments in Bács County and Iowa made itself felt to a great extent, too.

The introduction of the first Martonvásár SC hybrids coincided with the beginning of the intensive period of Hungarian maize production. The application of a higher dose of fertilizer per unit area, the increased application of herbicides and the use of more up-to-date seeders meant that a higher plant density per hectare was possible, and this was also neces-



sary, because of increased production costs. The use of combine harvesters became general within a few years. The importance of stalk strength became decisive. A shortage of combines made the harvest period considerably longer and this fact led to a higher percentage of broken stalks. So it can be said that the Hungarian hybrids, bred partly from Hungarian genotypes, became less and less suited to up-to-date technology. (This is quite understandable considering that the Hungarian-bred varieties were intended for extensive growing conditions, manual labour, etc. Since the individual productivity and the strength of root and stalk are generally in inverse proportion to each other, the genes determining root and stalk strength almost totally disappeared from the Hungarian-bred varieties.) By the end of the 1970s the line No. 156, evolved from the epoch-making Mindszentspusztai yellow dent variety, was almost completely eliminated from Hungarian grain maize production, together with the other Hungarian lines. This phenomenon both challenged and confirmed a number of previously held views on breeding and testing methods. At the same time it called attention to the fact that further changes are needed in maize breeding in Hungary. Before discussing these problems, let us say a few words about practical maize breeding apart from that in Martonvásár.

In the 1960s, besides the maize breeding in Martonvásár, which was financed partly from the research fund of the Hungarian Academy of Sciences and partly from the profits of seed production, practical maize breeding was carried out at the Szeged and Szarvas research institutes of the Ministry of Food and Agriculture and at the Agricultural University of Keszthely. A certain amount of methodological research was conducted at the Agricultural Universities in Gödöllő, Debrecen and Mosonmagyaróvár. The most significant results were achieved in Gödöllő by Professor Andor Bálint and his team. Very useful work was also done in the Agrobotanical Institute in Tápíószele under the leadership of Dr. Andor Jánossy. The results were utilized mostly in the research programmes of Keszthely and Szeged. The maize researchers from Mosonmagyaróvár and Lovászpátona were transferred to the university of Keszthely at the beginning of the 1960s. Among the research units of the Ministry of Food and Agriculture it was chiefly in Keszthely that a staff was developed for breeding early and very early hybrids. However, significant work was also devoted to medium and medium-late silage maize breeding by the leader of the team, László Berzsenyi-Janossits. The Keszthelyi 22 DC hybrid was a popular silage maize under cultivation for almost two decades. As in the silage maize hybrid Mv 26, which was also widely grown, lines evolved from the Mindszentspusztai white flint variety were of decisive importance in the hybrid K 22. One of the lines of Szarvasi DC 590, which was registered in 1969, was of the same origin. (With reference to the present situation, the best line evolved from Mindszentspusztai white flint was still being used with success in the hybrid Szegedi DC 538, registered in 1980.)

Although K 22 (DC) satisfied important production requirements, it was not this hybrid that represented the most characteristic contribution of the ministerial institutes to Hungarian maize breeding. The results achieved not only with Mv 40 but with Szeged 71 (DC) too, had already shown that growing late maize was not necessarily the source of the highest profit in maize production, even though the yield per hectare was the highest. By the end of the 1960s the higher water content, greater susceptibility to ear rot and late harvest, which made it extremely unfavourable as a forecrop, convinced many farmers, even those farming on areas very favourable for maize production, of the advisability of producing hybrids with a shorter vegetation. In Keszthely the Georgikon hybrids initiated a period during which valuable early hybrids were registered and introduced into commercial production. Besides the Hungarian inbred lines, public lines from the U.S.A., such as A90, W153R, Oh43, A632, etc., which had the qualities (stalk strength, prolificacy, adaptability, etc.) missing from the Hungarian lines or present only to a moderate extent, played a greater and greater role. Keszthely SC 360, Szarvasi SC 363 and Mv SC 380 from Martonvásár came into existence in this way. They became the most popular early hybrids in the 1970s.

At the end of the 1960s it became possible to utilize the results of Yugoslavian breeders in Hungary. They were in direct contact with the universities and private firms of the U.S.A., so they obtained public lines and hybrids regularly after World War II. It is obvious that the testing in Hungary of hybrids from the four Yugoslavian institutes (Zemun Polje, Zagreb, Novi Sad, and Osiek) hastened the process resulting in the breeding of up-to-date Hungarian hybrids and the testing and introduction of modern foreign hybrids (primarily from the U.S.A. and France) into Hungary. To start with, hybrids from Belgrade (Zemun Polje) and Novi Sad introduced some novelty into the medium-late and late FAO maturity groups (ZP-SK6, NS-SK70, etc.). Later, hybrids from Osiek and Zagreb became important in the earlier FAO maturity groups (OS-SK 218, Bc-SK 66-25, Bc MSC 418, etc.).

The demand for more up-to-date maize production, the personal experience gained abroad and the agronomic traits of the hybrids tested in Hungary, encouraged plant breeders in Keszthely to test the new combinations and experimental hybrids at a higher plant density



per hectare. When breeding earlier hybrids, excellent root and stalk strength became more and more important. In short: the production of hybrids which could be harvested by machine, even in the mature state, with minimal loss, became a justifiable and more and more urgent requirement. The recognition of this fact perhaps induced us to modify the breeding and testing programme in Keszthely somewhat earlier than was done in other institutes, and this indisputably gave us a definite advantage.

An important change in the use of T-cms in Hungary was brought about by the *Helminthosporium maydis* epidemic in the U.S.A. in 1970. It is to the credit of the breeders in Martonvásár that they used T-cms to achieve reliable seed production of the most important Hungarian hybrids by the end of the 1960s. The male sterile analogues of Mv 59 and Mv DC 602—just to mention the two most important hybrids—made it possible to produce genetically pure seed successfully, reliably and profitably. Based on the experiences in the U.S.A. the use of T-cms was officially prohibited in Hungary, too. To some minds, this was an over-cautious measure, but instead of lamenting, breeders promptly focussed their attention on non-T sources of male sterility. The international collaboration was again exemplary, and within 1 or 2 years each maize breeder working in this field received samples of the non-T cms sources available at that time. The Krasnodar Agricultural Research Institute, Cornell University, Ithaca, and the University of Illinois, Urbana, must be mentioned among the many institutes which offered their assistance. Important results were attained in the utilization and research of non-T-type cms analogues in the Szeged programme. These results were mainly due to the research carried out by Dr. László Kálmán. (In the 1981/82 season non-T-type cms analogues were sown on about 10% of the seed production area in Hungary.)

Summarizing the history of hybrid maize breeding it can be said that Rudolf Fleischmann and Endre Pap did pioneer work, not only in Hungary but in Europe as well, by producing inbred lines and by utilizing the heterosis effect in practice. László Berzsenyi-Janosits achieved long-standing results in breeding variety hybrids. The generations following them, particularly in Martonvásár, achieved important results chiefly in the production and propagation of SC hybrids; the researchers employed in the ministerial research programme in Keszthely, Szeged and Szarvas produced important results in breeding hybrids with short vegetation periods and better stalk strength.

For my part, I consider it no less important that Hungarian maize breeding, inbred maintenance, basic material multiplication and commercial seed production were ready for the adaptation and introduction of the most modern foreign hybrids by the end of the 1970s. This process was initiated in great measure by the production systems and it is maintained by the constant renewal of their demands. Even 10 years ago it was not the interests of narrow groups but those of the whole agricultural community which characterized this work, and it is still the needs of the national economy which guide it. This train of thought will perhaps provide the best bridge to take us from the past to the present.

### The present situation

The situation in maize breeding at the beginning of the 1980s is closely linked with the changes which took place in the previous decade in the organization of and conditions for maize breeding and production. The integrated, industrial-scale system of maize production was built up by the end of the 1970s. The main reason for establishing the production systems was to create the optimum harmony between all the production factors. The permanent development and maintenance of these factors was regarded as a further task. The changes in the biological background have brought the most striking success. Besides using the excellent Hungarian hybrids, which had good stalk strength, they did not hesitate to import hybrids from abroad, which were genetically different and often more valuable to farmers than some of the Hungarian ones, and to adapt them in cooperation with the relevant institutes. It soon became evident that the relatively modest additional costs of adaptation are reliably and economically counterbalanced by their higher yield. The well-known fact that most hybrids bred in North America and of suitable maturity for our conditions could be grown successfully in Hungary was also confirmed in practice. The best of these soon became leading hybrids. It was mainly American firms such as Northrup King and DeKalb which had a tradition of trade with Europe, and the most famous French firm of the period, Mais Angevan, which found the Hungarian market mutually useful.

The introduction of foreign hybrids was followed by the development of inbred maintenance breeding, parent and commercial seed production in Hungary, since the import of commercial seed was often rather uncertain. The hybrids NK-PX 50 and NK-PX 442 were among the first so-called cooperated foreign hybrids, whose seed was produced on 600 hectares



in 1973 mainly at the initiative of the Corn Production System. Although seed had been produced in cooperation with Yugoslav institutes at Zemun Polje, Novi Sad and Zagreb since 1969, it was mainly the American hybrids that gained a stable foothold.

But let us return briefly to maize breeding in a closer sense. In Martonvásár the breeding staff has been active for three decades. After the necessary development, 7-8 researchers are involved with practical breeding and inbred maintenance. Since the uniting of the Keszthely and Szeged programmes, 9-10 researchers have been carrying out similar work in applied research. The Táplánszentkereszt research station attached to the Cereal Research Institute plays a very important part in this work. The breeding and maintenance of very early and early hybrids is done here, including the majority of the institute's work in the GDR-Poland-Hungary cooperation, under the leadership of Zoltán Pintér. At the Seed Production and Marketing Company (the legal successor of the Irrigation Research Institute at Szarvas), the three agricultural universities, and the Agrobotanical Centre of the Plant Production and Qualifying Institute in Tápíószele there is an average of one researcher dealing with practical breeding, or with methodological research where the results can be utilized in breeding directly. Consequently, a total of 21-23 people are involved with maize breeding and variety maintenance in Hungary. A further 7-9 researchers working at the above institutes do physiological, pathological and cytological research on maize. Thus, the number of Hungarian scientists working on maize research does not exceed 30. (It is fortunate that three-quarters of these are at their most creative age and they show no lack of diligence, devotion and talent.)

On the other hand, I feel it is unfortunate that post-graduate training is often only possible at the expense of creative work. Nor can we boast that they all speak English, the "international" language of maize. Only about half of the researchers have an active knowledge of English, though most of them have a passive knowledge. However, if we compare the situation today with that existing 10-15 years ago, we will find a considerable improvement. The most recent regulations will further improve the situation, as they provide for post-graduate training to be financed from central funds. It must be mentioned here that the post-graduate training carried out at the Agricultural University in Gödöllő cannot be over-estimated, as very few graduates in biology and genetics just happen to take up maize breeding. Young researchers undergo intensive training in genetics and breeding at the Agricultural University of Gödöllő as the students of Dr. Andor Bálint. The success of him and his colleagues in post-graduate training is indisputable.

There are thus approximately 30 research workers in Hungary dealing with maize breeding, who are faced with the task of developing hybrids as good or better than the foreign ones, despite the fact that most foreign maize breeding stations in the temperate zone are incomparably better equipped with laboratory instruments and farm machinery. The fact that not everything depends on the number of breeders and equipment is proved by numerous examples. However, now that breeders in the temperate zone all use similar or identical source material and inbred lines, the time needed for selecting the best combinations from the infinite number of combinations is in direct correlation with the hours available for practical breeding and the number of trial plots. If this aspect is taken into closer consideration, the mere existence of the present Hungarian hybrids and experimental hybrids is astonishing. Why is this? The number of practical breeders in Europe is about 170. For the U.S.A. this figure is about 500, naturally including both the state institutes and the private companies as well. According to the official, very progressive Hungarian variety policy, if the partners conclude a mutually advantageous licence contract including parent seed multiplication and commercial production, and—in certain cases—inbred maintenance as well, then any hybrid developed anywhere from California to Krasnodar and from Rome to Hannover which gives reliably better results than the standards in the state trials is given the green light for introduction into Hungarian maize production. Under such circumstances Hungarian maize breeders have to compete with practically the whole world. There is, however, one sure winner in this competition, and that is Hungarian agriculture.

It can be said without any prejudice that even the fact that we are able to grasp the results of this rapidly developing science, to apply its best methods and to make the recent results from abroad available to farmers in Hungary without delay is in itself an achievement. If in addition Hungarian breeders are capable of producing practical breeding results, this deserves special attention and appreciation.

There is one thing, however, that Hungarian maize breeding can surely be proud of, namely, the maize breeding cooperation between the GDR and Hungary, started in 1965, the results of which have had a decisive influence on the very early and early silage maize production of the GDR, Poland and Hungary since the mid-seventies. (In 1973 the bilateral cooperation became trilateral when the Polish breeders joined the cooperation.) The joint



BEKE and BEMA hybrids proved to be excellent in the above countries compared to other European hybrids in similar maturity groups and for similar purposes. In my opinion, one reason of this success lies in the fact that the capacity of the nurseries in Bernburg, Kobierzyce, Smolice and Martonvásár together reached the minimum required to create adequate facilities for productive work. Besides the ideal personal contacts, the other reason is, that the breeding material used chiefly in the GDR, and the French yellow flint breeding material show excellent combining ability with the Hungarian and foreign dent inbreds. (However, it should be mentioned that there is some stagnation in very early grain and silage maize breeding because of the difficulties in achieving profitable seed production. In this field the conditions available to West European breeders are more favourable. A substantial improvement cannot be expected until there is a change in the price construction. This must depend on the profitability of hybrid production and—to a certain extent—on the value of the hybrid.)

Breeding for quality faces similar problems. The success in the production of high lysine maize is promising. The fact that it has proved possible to produce short season, healthy experimental hybrids is especially positive. However, there seems little chance at the moment of compensating for the 10–15% yield decrease associated with better feed quality by means of a higher price. The consequence of these problems cannot be avoided: in the breeding programmes the capacity available for objectives which will not give a profit within a short or medium period has been reduced. I know this is not a wise decision, and that we should be working for tomorrow, not for today, but the restricted capacity sometimes forces us to change our principles.

There is nothing to be ashamed of when we admit that Hungarian hybrid maize breeding was based in part on foreign research results right from the beginning. (In the make-up of Mv 1 DC, for instance, three American public lines took part.) But for our present work it must be taken into account that in the U.S.A., which is the most significant of our foreign cooperators, considerable changes have taken place in research in genetics, physiology and breeding. The essence of these changes is as follows. The development of university facilities was unable either to keep abreast of the increasing demands or to counterbalance the effects of inflation. Another reason for the change, as far as I can see, is that hybrid maize breeding, seed production and seed marketing have become very profitable activities. There is sharp competition between the private companies. Besides the open pedigree lines, privately owned inbreds or by researchers cooperating with the owner, subject to strict restrictions. Since the middle of the 1970s this process has accelerated. Today the original privately owned lines, and the sister-lines evolved from university lines and subject to exclusive utilization rights, dominate in those parts of the Corn Belt which are climatically similar to Hungary.

These facts make it quite clear what cooperating partners must be sought in Hungarian maize breeding and in the application of research results. It is an imperative to maintain the mutually advantageous connection with the foreign state institutions; in fact, these should be developed wherever possible. In my opinion, however, the most important source of further development is fruitful cooperation with firms which are well endowed with capital and brains.

The present situation of maize breeding is demonstrated in Table 1, which contains the genetic base of the hybrids on the basis of seed requirements. It is evident that the germplasm has been enhanced during the past 15 years. It is also obvious that the number and weight of the private lines from the U.S.A. has increased. This is in agreement with the tendency in other modern maize producing states. In my opinion, it is important to note that the significance of sister-lines has also increased. This proves the success of the back-cross, recurrent and pedigree selection methods. To my mind, an improved recovered line is no less of a breeding achievement than the selection of a new line from an elite population.

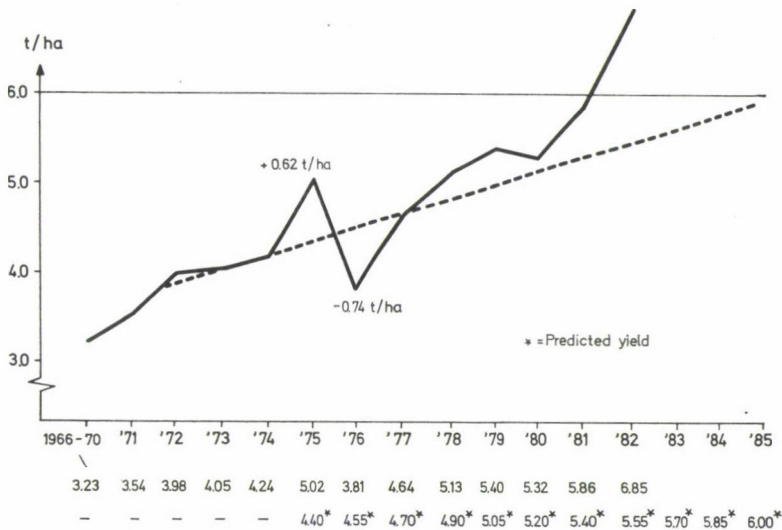
Another problem facing maize breeding at present is the amount of capacity which should be devoted to silage maize breeding. The justification for such work is no longer in any doubt, but the question still remains: if the capacity is limited, which programme should have priority. The least we can do is to choose from the most up-to-date hybrids available, those which are the most suitable for silage maize production, since it has been proved that there are genetically determined differences in quality and digestibility between the hybrids. Furthermore, the total dry matter yield of hybrids with the same grain yield is not the same. These main aspects should determine the objectives of silage maize production and breeding nowadays.

The efficiency of Hungarian maize breeding and adaptation is also shown by the fact that the maturity of the hybrids in production decreased by approx. 10% between 1973 and 1983 (calculated on the basis of the weighted mean of the seed used), coinciding with a dynamic increase in average yields, although with a modest decrease in the production area. There is no need to provide special proof of what an energy-saving and quality-improving

**Table 1**  
*Genetic base of maize in Hungary in 1968, 1973, 1978 and 1983*

%	1968		1973		1978		1983	
	No. of inbred	% of lines	No. of inbred	% of lines	No. of inbred	% of lines	No. of inbred	% of lines
over 5.00	6	73.4	5	65.6	5	50.8	5	67.5
2.00-4.99	8	24.0	6	16.6	9	23.5	5	15.8
1.00-1.99	1	1.4	9	12.6	10	12.9	6	8.5
under 1.00	3	1.2	10	5.2	25	12.8	24	8.2
Total	18	100.0	30	100.0	49	100.0	50	100.0
Approx. 75%	6	74.3	8	75.7	14	73.4	7	75.7
The 3 most widely used genotypes	A	18.4	A	21.2	A	15.3	A	28.0
	B	14.8	B	14.4	B	12.1	B	15.6
	C	13.6	C	12.8	C	10.0	C	11.6
Total		46.8		48.4		37.4		55.2
Total number and % of sisterlines of A, B and C		—		—	2	0.7	26	17.5
Total number and % of all sisterlines		—		—		—	43	21.3

step this is. But on the basis of Fig. 1 it can be seen that the climate is also of utmost importance (see the years 1975, 1976, 1981 and 1982). The question thus arises: how can Hungarian maize breeding create the necessary conditions to ensure that the hybrids and the seed should not be limiting factors to a further yield increase in the future either? What can we do to enhance the germplasm, to test the most up-to-date foreign experimental hybrids, and to be able to apply the most modern breeding methods? I will try to answer these questions in the last section of my paper.



**Fig. 1.** Grain yield of maize in Hungary



### The future of Hungarian hybrid maize breeding

Future tasks must be considered on the basis of the present situation. Plant production is the most important branch of agriculture in Hungary, and the prominence of maize production is indisputable. No significant change in the number or composition of the livestock can be expected. The export of animal products is a question of basic importance for the state economy, and there is also a modest export of maize (approx. half a million tons). The state economic plan assumes that the genetic potential of maize will continue to increase as it has up till now. But it also expects quite justifiably that the utilization of the genetic potential which depends on human factors should increase. By the end of the 20th century a yield average of 7.5–7.6 t/ha is not unrealistic. But to accomplish this plan a full scale development of Hungarian hybrid maize breeding is indispensable.

In my opinion, it is very important to increase the basic research capacity. This could be accomplished in two mutually complementary ways. The team working on basic maize research at the Agricultural Research Institute of the Hungarian Academy of Sciences in Martonvásár should definitely be strengthened. The research facilities already available make this a rational step, which is also supported by tradition. The other way is to develop basic and supporting research at the universities. (In my opinion, it is against human nature for the knowledge of an excellent, creative teacher to be based exclusively on special literature.) This is emphasized by the fact that, apart from the Biological Research Centre in Szeged, there is no forum other than the universities for discussing and solving the problems of maize research with the highly trained specialists of related branches of science.

The general level of special education (thorough knowledge of basic sciences, knowledge of foreign languages, organizing ability, etc.) of those who work on practical research and maize breeding, should be raised. Biologists and geneticists with non-agricultural academic qualifications should be encouraged to work in greater numbers on maize breeding research. There should be more opportunity than at present for young researchers to spend a certain time in developed foreign countries in order to learn the most modern methods and to widen their experience.

In order to close or narrow the gap between Hungarian maize breeding and that of the most advanced countries in this field, the primary need is to urgently provide all the material facilities badly needed nowadays. There is an imperative need for changes, which will require a considerable allocation of funds, due to the following factors: the mechanization of plot trials is very low; there is a considerable lack of modern laboratory equipment; the alarming backwardness in recording, evaluating and compiling data; and the primitive processing and storage conditions for breeding and experimental stocks. In this connection, I see the greatest contradiction in the fact that, while the technical conditions of production have developed rapidly over the last decade, attaining a very high level on many farms, the funding of research has improved only modestly, and even adaptation ability has sometimes been questioned.

In my opinion, it has become quite obvious that it is impossible to create harmony between the biological and other factors of production without a significant improvement in Hungarian research conditions, particularly as regards finance. This situation is a serious threat to a further increase in yield averages. The 1% increase in yield averages which is attributed to genetics in countries in the temperate zone, amounts to more than a quarter of a billion forints per year in Hungary. This sum is at risk if the development of research does not keep abreast with general technical development. (Research should preferably be developed at a higher rate than the latter.)

The objectives of maize breeding have not changed essentially. Hybrids must be produced which yield more than those currently under cultivation, or which have the same genetic potential but better yield stability and lower production costs. These latter aspects modify breeding objectives to a certain extent. The following points can be briefly mentioned:

- the hybrids of the future should give reliably good yields even under less favourable environments, whereas they should respond to favourable conditions with a high yield;
- they should be produced economically with the application of less N-fertilizer;
- their germination and seedling vigour should enable optimal plant density even in seed-beds which are less favourable, due to energy-saving tillage;
- they should be more tolerant to both drought and high temperature than the present hybrids;
- their resistance to pests and diseases should be based on a more diverse germplasm than that of the present hybrids;



- the amino acid composition of maize hybrids with normal endosperms should be more favourable; the proportion of lysine should improve without a decrease in yielding ability.

This list only includes the most characteristic changes, but these are enough to require a certain modification in the choice of breeding sources and breeding methodology. According to a number of well-known specialists, if the total number of maize germplasms available in the world are compared to a floating iceberg, the varieties and hybrids used at present only represent the tip of the iceberg. More accurate estimations lead to the conclusion that only about 2% of the known germplasm has so far been used by breeders and farmers. Needless to say, this is probably the best 2%. But this is only a probability, because we know little or nothing about the genetic and agronomic traits of the other 98%. The reason for this is extremely simple. A great deal of money, human energy and time would be needed to describe and test this huge material and to prepare its genetic codes. At present this is an unworkable proposition in Hungary. The trouble is, however, that this work is difficult to carry out in other parts of the world, too. Since 1965 the funding of research at public expense has been on the decrease in the U.S.A. This could lead to considerable negative changes in the utilization of the results of the international connections built up during the past four decades. The possibility of discovering and applying the genetic source materials hitherto unused is gradually becoming the exclusive precinct of seed firms which are well provided with capital and carry out extensive agro-genetic research in the U.S.A.

It seems to me that one realistic way of achieving the modified breeding objectives outlined above would be to enhance the germplasm by introducing exotic materials. Although these breeding materials are difficult and circumstantial to handle and to adapt, they are suitable for improving yielding ability, resistance and feed quality. Successful adaptation and utilization would require several decades of work by highly qualified specialists, and a considerable allocation of funds. Naturally, Hungarian possibilities are limited primarily for the latter reason. The situation is somewhat similar at the universities of the U.S.A., although to a lesser extent. Thus the utilization of this avenue must rely on prosperous private firms.

Many people feel that genetic advance will slow down within one or two decades and then cease completely, because of the known genetic materials and classical breeding methods. (This is not in contradiction with the fact that the present germplasm will make an average 1% advance per year possible for 15–20 years to come.) If this predicted stagnation is to be avoided it is now that breeding methods must be elaborated and gradually developed for use at the end of the 20th century. Most people think that one way out is biotechnology and this is gradually becoming the "official" opinion, too. The application of biotechnology again needs well-trained scientists, and to some extent more expensive equipment, etc. The experience gained so far indicates that only long-term research can lead to practical results. But even before this stage is reached it should lead to improved knowledge on the physiological and biochemical processes of the plant, and it will enhance the chances of success in classical breeding. What place will biotechnology and genetic engineering have in Hungarian maize breeding? At present one maize researcher is dealing with this modern method, but not on a full-time basis. Soon another researcher will be joining him. The first objective is for these researchers to be able to understand the results attained in the main "maize research" countries and to apply them successfully as far as is possible in Hungary. Recent international experience shows that private firms are taking a more and more active part in this research area and are incorporating this new method into their long-term plans. (They are encouraged to do so by the lack of state research capacity, besides their own well-considered interests.)

If, in addition to the three large areas mentioned above, namely enhancing germplasm, using exotic materials and developing biotechnology, it is also considered that private firms are able, for known reasons, to do increasingly more than the universities even in the field of recording, evaluating and compiling data, there is no escaping the following conclusion: if the standard of maize research and production is to be maintained or even raised, mutually advantageous cooperation must be established on a contract basis with the private firms endowed with considerable research capacity in the leading maize research state of the temperate zone, the US. In addition, contacts must be retained with the publicly financed research institutions of North America. In this field purely business interests do not dominate the scene completely: the traditionally international character of agricultural research results is still strong, since the result of this work is our daily bread, badly needed in the developing countries to still hunger and to save millions from starvation.

If more funds were allocated for state research in the U.S.A., Hungarian maize breeding research would be somewhat less dependent on private firms. But this dependency can also be diminished through the established connections with European countries, both from Comecon and the Common Market, because the indirect flow of overseas research results is

an appreciable source of development. But if the policy of retardation continues to predominate in the field of research in the U.S.A. in the future, not only will the results of supporting and applied research become monopolistic, but the most talented scientists will be working for private firms, and within 10-20 years the training of researchers will itself be seriously endangered. This could lead to an extremely harmful vicious circle. We can only hope that this will not happen, because the individuals and bodies responsible for making decisions will support development due to the undeniable benefits of plant breeding. Numerous surveys in the U.S.A. prove that 35-55% of the sums invested in agricultural research is repaid yearly (!). It must be clear to all concerned that the foundations of the research results to be achieved at the end of the 20th century and beyond must be laid now.

What is to be expected and what must be done in the field of maize breeding research in Hungary? In my opinion, the tasks facing researchers today and in the years to come are the following:

- a) more, better-trained staff must be employed in basic research;
- b) there is an imperative need to raise the standard of training for specialists in applied research;
- c) the capacity of breeding nurseries must be increased: breeding material of greater diversity and a larger number of plots should be used;
- d) work with modern plot machines, laboratory equipment and computers must become natural, together with the modern field laboratories and stores for breeding stocks which make effective research possible;
- e) the financial resources and possibilities available at present in the plant breeding institutes should be used in such a way that plant breeding and variety maintenance are given unquestioned priority;
- f) there must always be a willingness to learn and profit from the results attained in any country of the world and to apply them without delay if they can be of use in Hungary, whether they are inbred lines, hybrids or breeding methods;
- g) Hungarian maize breeding work should always produce a sufficient quantity of unalienable results of its own, so that, if the need arises, it can make modern biological production factors available to farmers without any significant decrease in yield.

The above objectives and conditions will only materialize if there is a more effective coordination of domestic maize research. It is indispensable to utilize the present tools of maize breeding more effectively and to bring about a less ambiguous funding of research. There is also a need for part of the profits of the farms to find their way into the development of research. (A good example of this is the constructive connection between the farm and institute of the Hungarian Academy of Sciences in Martonvásár.)

It is indispensable to develop the common interests of the individual and of the team as a whole: financial appreciation should be in harmony with the results achieved. In my opinion, this could all be achieved without risking the necessity and conditions for healthy competition between the applied research groups in Hungary.

In the future there will be less striking success in maize breeding than in the past three decades. The starting-point is also incomparably more advanced than it was 30-35 years ago. The economic situation in Hungary and in other countries does not make non-essential development possible. But the groundless retardation of research development is equivalent to a voluntary and irrational abnegation of the results which could be achieved in the future. Every decision-maker should be aware of this fact, as I am sure they are. Consequently, we can face the future with optimism, ready, if need be, to convince others with our arguments.

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## CHRONICA

### GARDENING IN HUNGARY IN THE DAYS OF TURKISH RULE, IN THE 16TH CENTURY

In memory of SÁNDOR TAKÁTS, on the 5th anniversary of his death

*Sándor Takáts* (1860-1932), historian, was a pioneer in the Hungarian history of culture and economics. He obtained a teacher's diploma of history and Latin at the Budapest university, and in 1881 entered the Piarist Order. In 1898 the Hungarian government sent him to Vienna to carry out investigations into archivalia. He worked in the archives of Munich, Nuremberg, Augsburg, Passau, Brno, Graz and Laibach.

He was particularly interested in the period of Turkish occupation. In possession of a vast amount of archival data, he disputed many points of the old chronicles. He pointed out that Hungary had hardly ever a more enthusiastic and nationalistic period than in the 16th century. He gave evidence of beneficial innovations by the Turkish population of Hungary, and did much to increase mutual understanding.

With this many-sided activity, *Sándor Takáts* did much to create the history of economics and culture, which had previously been a rather neglected sphere of historiography, and popularize it in Hungary. It is with this humble work that we commemorate the indefatigable researcher, the renowned historian, the unprejudiced Piarist teacher and member of the Hungarian Academy of Sciences, on the 50th anniversary of his death.

#### 1. Gardening in the 16th century

This era was the period of Hungary's being torn into three parts. In 1526, on the field of Mohács the independent Hungarian state came to its end. As seen on the annexed map, the Hungarian kingdom was confined to Western Transdanubia and the northern medium-high mountains. The central part of the country lay under Turkish rule, while in the East the Transylvania Principality fought to survive.

In that period, Hungary was truly called a land of fear. Turkish and Hungarian troupes raced about the country. Highway-men and brigands hunted for travellers. The roads were indescribably bad, even the next village could not be reached without breaking a cart wheel; and the low-lands were covered by moors, bogs and reeds. The stagnant waters polluted the air. Travellers, delegates and soldiers who came to Hungary at that time unequivocally wrote that the water and air of Hungary were pestilential.

According to the old, documentary testimony, horticulture in Hungary was on a very high level in that period. Since gardening is not only the cradle but also the thermometer of culture, we can safely speak of a progressive and strengthening culture during the 16th century. Eternal humanity, beauty, the love for flowers, all those pleasures offered by the garden, could never be killed in the souls even by the most savage conflict. Those who dealt in death, thought in terms of life; and even amidst slaughter and destruction, they longed for peaceful occupations.

All this is partly due to the behaviour of the Turkish conquerors. The communication between Hungarians and Turks had beneficial effects on horticulture in Hungary. Hungary became a mediator between East and West as regards fruit and flower cultivation. The letters of Turks living in Hungary, and Turkish travellers, always spoke with great enthusiasm of the vineyards and orchards of the occupied Hungarian towns. The savoury fruits, sweet grapes and fiery wines of Pécs or Eger were described in the highest rhetoric. All this is proved by

masses of official documents. Turks living in Hungary devoted themselves to gardening. They introduced a wide range of fruit species, flowers and commercial crops.

When speaking of horticulture during the Turkish occupation of Hungary, we must not think of the Italian ornamental gardens of the Renaissance, nor of the Italian style gardens of King Mátyás and Archbishop János Vitéz. The economic conditions of the 16th century did not favour the spending of much money. At a time of continuous fightings, who could have afforded to embellish his garden with fountains, waterfalls, statues, loggias? Ornamental gardening was therefore replaced by useful gardening. In the 16th century, both the landlords and the serfs made every effort to produce fruits and vegetables of the highest possible quality. In their love of flowers, they planted them also within their vegetable gardens. There were but very few aristocrats in 16th century Hungary who could boast of having separate flower gardens.

Numerous data prove that the 16th century was the golden age of Hungarian horticulture. We have never reaped as much honour with the produce of our gardens as in that century. The leading role was played by the noble ladies, quite understandably, as their lords of the 16th century spent nearly all their time in border fortresses, fighting the Turks or taking part at meetings on national affairs. They mostly returned home only in the time of harvest, when — as a rule of the borders — conflict was suspended. Thus, the management of farming, household and gardening depended upon the lady of the house.

Those noble ladies are worth being remembered for their farms and gardens; "they were flaming torches lighting not only the past but providing a guiding light for the future, too."

## 2. Gardens of aristocrats in the 16th century

We shall subsequently describe the gardens and show the gardening activities of three aristocratic families in the 16th century, on the basis of data found in Sándor Takáts' books.

### a) The Csurgó garden

The favourite residence of Katalin Pemflinger, wife to *Bálint Enyingi Török* (?–1551), called "leaena lutterana" in the Szerémi Chronicle, was Csurgó. It was there that she very willingly managed the estate. In the big garden of Csurgó, she played with her two sons; and found comfort in her sorrow over her husband, held in the Seven-Tower Prison of Istanbul.

This garden of Csurgó is the only one of which detailed description has been left to us from those days. In view of the history of culture in Hungary, this description is of outstanding importance, because we can learn from it about what were to be found then in a Hungarian garden. You can imagine how large that garden was, when considering that two cadastral yokes were planted with melon only. Vegetables occupied an area of several cad. yokes; there were 21 fields of onion, 1 cad. yoke and 9 fields of tall-growing peas, 29 fields of chick-peas, 15 fields of lettuce, 4 fields of spinach, 2 fields of leeks, 2 fields of garlic, 2 fields of shallot; 43 seed cabbages, 7 cypresses, 200 bushes of picea, 88 trees of Persian and red clingstone peaches, 82 trees of Besztercei- and other plums, and 18 sour-cherry trees.

The beds were bordered by those sweet-smelling flowers much admired in Hungary; rosemary, sage, lavender, lily, hyssop, and rue. White lily seems to have been the favourite flower of the lady of the house, as 400 of them were found in her garden.

### b) The Sárvár garden

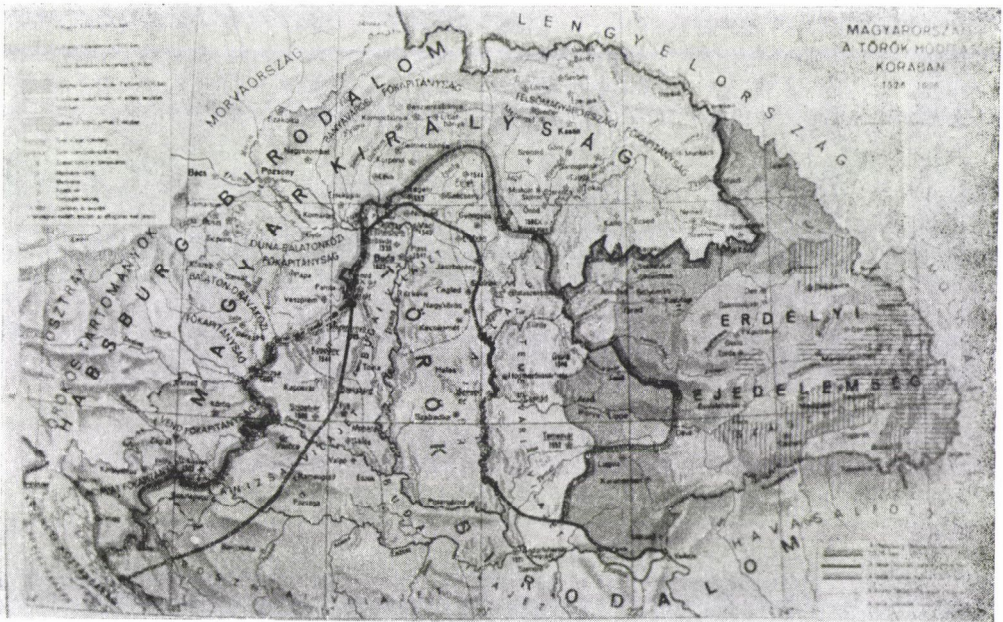
The first and most prominent gardeners of Hungary in the 16th century were *Tamás Nádasdy*, Palatine (1498–1562), and his wife *Orsika Kanizsay*. According to contemporary letters, their garden at Sárvár was the finest one in the country. The Palatine himself was the best gardener. Everything was done according to his instructions. He prescribed for his gardener how to manure the trees and treat them with grounds of wine so as to encourage larger fruits. The melons were protected against the cold, according to his directives. He even showed the gardener how to twist the stems of plums before they ripened.

As a Palatine (the highest administrative dignitary in Hungary before 1848) he received fruits from all parts of the country. If he found an excellent one he immediately arranged for a graft of the tree to be sent to him. Thus, *Nádasdy* planted trees of all the good fruit varieties in the country on his land, and supplied others with scions and grafts. He propagated for example, the plantation of muscat grapes. On 6 January 1551 Archbishop *Miklós Oláh* sent a letter by courier from Augsburg in which he requested apple, pear, peach and plum scions from *Nádasdy* for Queen Maria, with the note that the names should be written on





*Fig. 1. Portrait of Sándor Takáts (1860–1932)*



*Fig. 2. Hungary during the Turkish occupation*





Fig. 3. Portrait of Tamás Nádasdy (1498–1562)

them. To Countess Salm, *György Török* sent young trees from Nádasdy's garden. In 1558 *Margit Széchy* asked Nádasdy to give her scions. *Pál Bornemissza*, Bishop of Nyitra, asked Nádasdy's wife to send him fruit grafts. Many grafts planted in the garden of King *Ferdinánd I.* (1526–1564) also came from the Sárvár garden.

His duties often kept *Tamás Nádasdy* away from his home. On such occasions all tasks of gardening were placed in the hands of his kind-hearted wife, *Orsika Kanizsay*. This noble lady loved gardening perhaps even more passionately than did her husband. The Sárvár garden became famous even in remote countries, since Lady Nádasdy sent fruits of her garden not only to her husband but also to members of the royal family. Her husband wrote her more than once on the success of her muscadine, melon or cherry with the Queen.

It might be thought surprising that the Nádasdys already had ripe cherries in the first days of May, and had asparagus and green peas still much earlier. In the first half of June, Lady Nádasdy even sent ripe melons, white plums and apples to her husband. In 1549 she astonished *Katalin Frangepán* by sending her cabbages in May.

In 1562 *Tamás Nádasdy* died. His widow continued to send the wonderful fruits of the Sárvár garden to the royal court and to the aristocratic families. For example, when in 1566 the King was encamped at Győr with princes and aristocrats, Lady Nádasdy sent them delicious fruits. Her scribe *Ádám*, who gladly delivered these, reported to his mistress that the Emperor and the Princes welcomed her gifts. Count *Harrach*, one of the noblemen present in the camp, who was a passionate gardener and who had acquired many fruit-trees both from Hungary and from abroad, remarked — according to the scribe *Ádám* — that such excellent fruits as those of his mistress could be obtained neither in Hungary nor from abroad, neither from noblemen nor from others; "Such an excellent farmer is your Ladyship".

The gardener of the Sárvár garden is known: his name was *Estván Kerty* and he had much to do with the fame of the Palatine's garden. He was an educated man who could write well in Latin and Hungarian; and, because the famous Sárvár garden was visited by many noblemen and dignitaries of the Church, he did everything to live up to expectations. In 1552 he could safely write to his master: "The garden is in order. Even if the King came, he himself could only say that that is all right. The paths are clean, everything is in order. The son of Master *Lukács Székely* — he writes — is there every day and says that he has never seen a finer garden than this." This *Lukács Székely* was one of the most prominent connoisseurs of gardens in those days. An excellent peach variety was named after him. His garden was famous

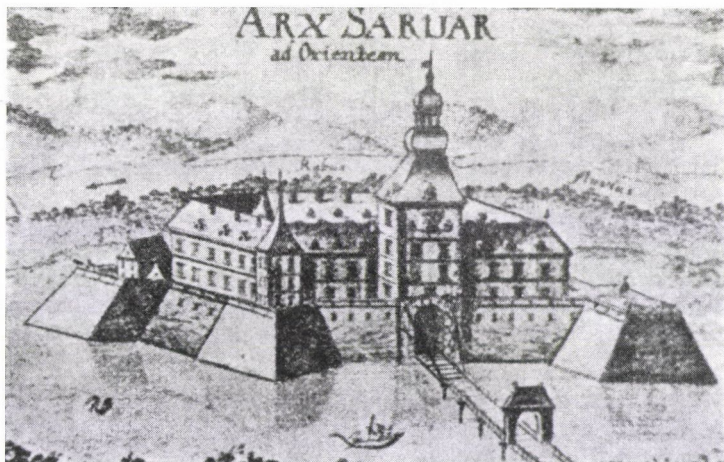


Fig. 4. Castle of Sárovar



Fig. 5. Hungarian fate. Xylograph from 1543

all over the country. The first apricot variety was found in the Sárovar garden. The plant had been introduced into Hungary by the Turks in the middle of the 16th century. The Nádasdys referred to it as "tengeribarack" (*Prunus armeniaca*). *Estván Kerty* naturally mentioned this rare tree also in his letters.

It was here, in the Sárovar garden that Nádosdy and his wife spent the best days of their lives, and here played the little *Ferenc Nádasdy* (1555–1604), later to become the brave military leader whom the Turks called, "Black Bey". He, too, took much care of the garden, and his gardener is likewise renowned. He also sent fine fruits and young fruit-trees to the Vienna Court. It is known, too, that the later Count *Ferenc Nádasdy* (1625–1671), Lord Chief Justice (the highest dignitary after the Palatine) paid great attention to his gardens. He had tulips, other rare flowers and shrubs sent to him from the Netherlands, and also found there a clever gardener for himself.

When, however, the Lord Chief Justice, as one of the leading members of the *Wesse-lényi* Conspiracy, was sentenced to death and decapitated, his properties were confiscated, after which the Sárovar garden soon fell into ruins. Its valuable plants were taken away.



Emperor *Lipót* (1658–1705) even had the rare animals removed from its forest-range. According to the contemporary documents, a few decades later the garden was mostly covered by grass and weeds.

c) *The gardens of Némétűjvár and Rohonc*

*Ferenc Batthyány* (1497–1566), a major landowner in Transdanubia, Governor of Croatia, founder of the fortunes of his family, was given the Némétűjvár estate in 1524. (Némétűjvár is today Güssing and belongs to Austria.) Although frugal, he was never sorry for spending money on farm investments, as a result of which his gardens and fish-ponds became famous everywhere.

His loving wife, *Kata Alsólivdvai Bánffy*, was a helpmate, adviser and all he could expect in managing the estate. She was one of the first noble ladies in Hungary, not only by origin but also for her education and character. At Némétűjvár and Rohonc (today: Rechnitz in Austria) everything connected with the estate and the household was almost her concern. She not only made arrangements and gave orders but did actual housework, too; spinning, making wreaths, sewing and weaving bone-lace with the girls educated in her court. When the weather turned warm she occupied herself as well in horticultural work. Once she wrote to her husband: "We have had much to do. We have tied up the wines and planted cabbages." On another occasion she gave account of having sown melons, distilled scents ("flower waters") and made various fruit preserves. She often surprised her acquaintances with delicious fruits, muscat- and other melons. Several times she sent fruit to the King and Queen, and even to the Archbishop of Esztergom.

When *Kata Bánffy* died, *Ferenc Batthyány* after a year of mourning, married *Kata Svetkovics*, an extremely thrifty woman, who, nevertheless, became one of the greatest benefactresses of her age. She, too, was willingly occupied with gardening. She had fine fruit, vegetable and flower gardens. She did the work of planting and sowing by herself and was proud when she could offer somebody the produce of her gardens.

After her husband's death, she managed her estates with great care, thanks to the assistance of a clever steward named *András Bagody*. It should be noted, that she employed



Fig. 6. Hungarian nobleman in the middle of the 16th century. Xylograph by Jobst Amman



vine-dressers from Tokay to work in her vineyards. From year to year, she added new grafts to her fruit-tree plantations. She even had a fine garden in a suburb of Vienna that she willed to her physician, the famous Pistalocius. It was the latter who converted the family *Batthyány* to the "new faith", the religion of the Reformed Church.

The Némétújvár and Rohonc estates — together with other landed properties — were inherited by *Boldizsár Batthyány* (1543–1590) and his wife *Dóra Zrínyi*, daughter of the hero of Szigetvár. *Boldizsár Batthyány* was one of the greatest Hungarians in the 16th century and an interesting figure in the Hungarian history of science and culture. He pursued studies in Paris, whence he returned with Huguenot ideas. He regularly had the newest French books sent to his library. Besides his native language, he spoke six others; and although he had his share in fights with the Turks, and was appointed Warden of the King for his military merits, he was deeply interested in science and horticulture. There were hardly any contemporary writers and scientists of note with whom he was not in constant correspondence. In the 1580's he promoted the valuable botanical work of *Clusius* (Charles l'Écluse), the famous botanist of the Netherlands, who spent a considerable time in his court. On the activity of the scientific circle of Némétújvár, two papers have been published by the author in this journal: 21, 1972: The Birth of Mycology. International scientific co-operation in the 16th century; 22, 1973: *Clusius Beythe*: *Stirpium Nomenclator Pannonicus*.

A further proof of *Boldizsár Batthyány's* love of horticulture is the fact that he released his high-born Turkish captive in return of Turkish flowers brought from the Sultan's garden. He was then requested to send tulips and other Turkish flowers abroad. For the learned doctor *Corvinus*, he had carnation roots sent to Vienna. In 1585 Count *Ruprecht Herberstein* wrote to him from Graz that the Turkish flower bulbs sent by *Batthyány* to his daughter had gone bad, so he asked for new ones and *Batthyány* fulfilled this request willingly. On 6 March 1587 *János Honelius* asked him to send rose of Jericho, double stock, tulip, narcissus and other rare bulbs to him in Marburg.

Although a large part of *Boldizsár Batthyány's* letters are lost for us, those still available testify that foreign gentlemen and scientists continually requested him to send them



Fig. 7. Yellow day lily (*Hemerocallis flava*) in *Clusius'* floristic work of Pannonia

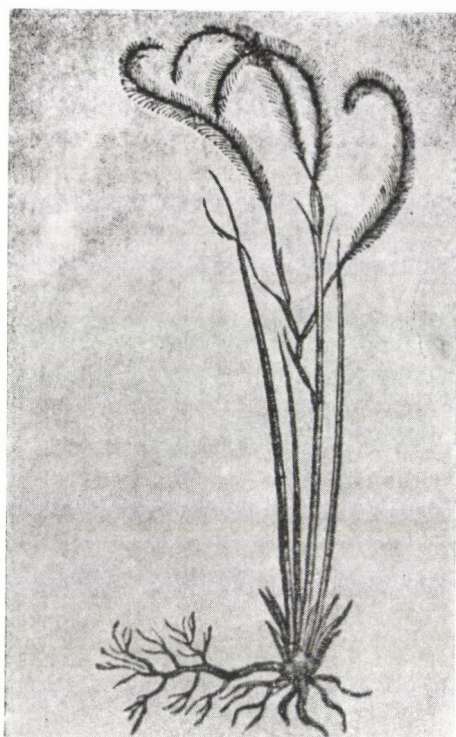


Fig. 8. Needlegrass (*Stipa Joannis*) in Clusius' floristic work of Pannonia

flowers and trees. Count *Nogarolla* asked in 1571 for melon and peach; and *Ozwaldt*, a dignitary of the Church from Voran, asked for pear scions. In 1573 King *Maximilian* (1564–1576) himself made the same request. *Elias Corvinus* thanked him for sending cling-stone peaches to him and to the Bishop of Würzburg. Among the documents of the Körmend Archives — *Sándor Takáts* writes — many similar letters of request are found.

When, at the age of 47, *Boldizsár Batthyány* died, all burdens of managing the large estates were laid on his wife, *Dóra Zrínyi*. She, though often in bad health, fulfilled this task with great pleasure and skill. It was gardening that she most enjoyed. She continually sent early and excellent fruit to her children and relatives. According to *Sándor Takáts*, "She was a simple, pure-minded, very active Hungarian lady".

She was sad to learn from her steward's letter of 1 June 1605 that at the time of the *Bocskai* uprising, her farm and gardens were destroyed. But this was already in the sad 17th century when many of our fine gardens went to ruin, later to give place to new ones, which then became tokens of a will to live and fondness of gardening. In that very 17th century, the first book on horticulture was published in Hungary.

P. HARGITA

#### References

- TAKÁTS, S. (1907): *Művelődéstörténeti Közlemények. Századok* (Cultural History Publications. Centuries). A Magyar Történelmi Társulat Közlönye. Budapest.
- TAKÁTS, S. (1914): *Régi magyar asszonyok* (Hungarian women of old times). "Élet" Irodalmi és Nyomdai Rt. Budapest, 1–341.
- TAKÁTS, S. (1915): *Rajzok a török világból* (Representation of the Turkish times). Magyar Tudományos Akadémia Kiadása. I. Budapest, 1–438.

- TAKÁTS, S. (year unknown): A török hódoltság korából. Rajzok a török világból. IV. befejező kötet (Of the Turkish occupation of Hungary. Representation of the Turkish times. IV. last volume). Genius kiadás Budapest, 1-570.
- TAKÁTS, S. (year unknown): Magyar nagyasszonyok (Noble ladies of Hungary). Genius kiadás, Budapest, 1-538.
- TAKÁTS, S. (year unknown): Szegény magyarok (Poor Hungarians). Genius kiadás. Budapest, 1-499.
- TAKÁTS, S. (year unknown): Régi idők — régi emberek (Old times — old-time people). 2nd edition. Athenaeum Irodalmi és Nyomdai Rt. Budapest, 1-471.
- TAKÁTS, S. (year unknown): Hangok a múltból (Voices from the past). Athenaeum Irodalmi és Nyomdai Rt. Budapest, 1-413.
- TAKÁTS, S. (1979): Buda két árulója (The two traitors of Buda). Szépirodalmi Könyvkiadó. Budapest, 1-338.
- TAKÁTS, S. (1979): Bajvívó magyarok (Hungarian champions). Móra Könyvkiadó. Budapest, 1-337.





## RECENSIONES

E. TÓTH—D. SURÁNYI: *Szilva (Plums)*. Mezőgazdasági Kiadó, Budapest, 1980. 428 p., 91 figs, 103 tables

In the first half of this century, and also in earlier periods, the plum was a much-liked, wide-spread fruit utilized in various ways in both Hungary and the neighbouring countries. However, in the second half of the century the number of plum-trees in Hungary was sharply reduced, involving a simultaneous decrease in the market supply of plums. In recent years, however, renewed interest has been shown in plums, so an improved quality and increased volume of plums and the further development of plum production are soon to be expected in Hungary.

Taking this into consideration, the timely publication reviewed below is to be welcomed; the structure, contents and up-to-date professional standard of the book supply valuable information, based on numerous original data and experimental results, for large-scale plum growers, the owners of small gardens, university students and scientific research workers alike. A vast amount of literature was used in writing the book: the references (pp. 401-418) include 269 works dated between 1882 and 1980, 137 of which were published in the seventies. The observations and experimental results of the two authors are represented by a total of 35 publications (15 and 20, respectively).

The clear text of the book is well complemented by the wisely chosen, conclusive documentation, which includes figures of a morphological, anatomical, systematical and physiological nature (drawings, photographs, column and circular diagrams) and various types of tables containing numerical data. The majority of the illustrations are original.

The structure of the book is logical and well-proportioned. Some chapters or parts of chapters were written in co-operation with Domokos Andor, Sándor Bognár, Sándor Brózik Jr., István Gergely, József Harsányi,

Mrs. Tamás Kállai, Gabriella Kovács and Endre Szücs.

In the Preface József Pekó speaks of the widespread cultivation and utilization of plums and emphasizes their importance in the public food supply both in the past and in the future. The book is then divided into 22 chapters broken down into sub-chapters, and occasionally to further sub-sections, according to need.

In the first chapter: Botanical Description, Geographic Distribution of Plums; Major Plum Species, a brief survey is given of the evolution of species and varieties in the genus *Prunus*, the history of plum cultivation, and the classification of the different species. In the sub-chapter Taxonomy of Plums the European, East Asian and North American species of *Prunus* are listed in three groups. The subsequent sub-chapter gives a short description of the above-mentioned species and varieties, illustrated with morphological photos, drawing of leaves and fruits, and tables of flower and fruit measurements. This chapter also deals with the major root-stock varieties used for plums.

The second chapter, covering nearly 60 pages, discusses the vegetative and reproductive characteristics of plums with the aid of tables containing numerous data from observations and measurements, and of a phylogenetic and phenological nature for the plum varieties studied; various diagrams help to clarify the questions of dormancy, forced dormancy, bud burst, leafing, shoot growth, and the colouring and falling of the leaves. Within the framework of the biology of flowering and fructification, the author deals with the course and date of flower bud differentiation, flower formation and the laws governing flower organization, in particular the conditions under which the androecium and gynoecium are formed, including photographs showing various irregularities. Many valuable observations are given to back up the detailed description of flowering and

pollination (open pollination, self-pollination, cross-pollination) in various plum varieties and the rather complex question of the setting, growth, development and ripening of the fruit. The last section of the chapter gives information on the productivity and yield of certain plum varieties and clones, how these are annually compared, and other related questions on the basis of specific data.

Chapter 3, entitled *The Economic Importance of Plums*, deals with economic, cultivation and biological aspects, calling attention among other things to the major nutrients and the number of calories found in 100 g of the ripe, fresh or processed fruit of several fruit species, to the qualitative and quantitative distribution of amino acids in two types of plum (a free-stone plum and a red plum), and to other valuable components and curative compounds.

Chapter 4 deals with the history of plum growing, referring to the written evidence of the nearly five thousand year past of this cultivated plant, including archeobotanical finds and the characteristics of the excavated remains of *Prunus* stones. The site, age, quantity, species and dimensions of major finds of *Prunus* stones in Central Europe are presented in tabular form, covering 3 pages. Mention is made in this chapter of plum growing in the 9th to 17th centuries, and of the history of plum growing in Hungary.

In the fifth chapter a survey is given of plum research in Hungary, emphasizing its importance and necessity. We learn that the pomologist Bereczki reported in some detail on the characteristics of as many as 136 plum varieties more than a hundred years ago. And a hundred years ago Staub prepared the first phytophenological map of Hungary, including many data on the flowering date of plums. Research continued and became more intensive, especially from the middle of this century, due primarily to the establishment and successful work of the Horticultural Research Institute.

In the sixth chapter, which deals with the situation of plum production, marketing and utilization, the plum cultivation of the world in general and of Europe and some European countries in particular is discussed, giving an insight into the cultivation, production area (including number of plum-trees), marketing and utilization of plums in Hungary.

Chapter 7, *A Brief Account of Plum Breeding*, deals with the objectives, methods and initial stock of plum breeding, the major problems of crossing and clone selection, and methods of introducing new species.

Chapter 8 supplies more than twenty pages of detailed information on plum varieties, listing not only registered varieties,

those with provisional propagation permits, those recommended for farm-scale variety trials and those in the process of being introduced, but also the new Yugoslav plum varieties and four new Romanian plum hybrids. The text is well illustrated with drawings of the shape and size of the fruit in certain plum varieties.

Very useful information and practical experience is summed up in Chapter 9, under the title *Propagation of Plums*, in which we first read about propagation from seed and learn that this method is used mainly in root-stock raising, and for breeding and research purposes. Vegetative propagation, which is generally carried out by grafting, and to a lesser extent through the development of accessory organs, is more widespread. Further questions discussed include root-stocks (seedling and clone stocks), the interaction between stock and scion, and finally the use of intergrafts.

In Chapter 10, *Ecological conditions required for plum growing*, a broad range of information is given on the light, heat and water requirements, the frost sensitivity and winter hardiness, the drought tolerance, the resistance to air currents and air pollution, and the optimum nutrient status of plums (i.e. the soil conditions at the growing site), with numerous examples, concrete data and observations to support it.

Chapter 11, *Establishment of Plum Orchards*, discusses how the plans should be prepared, various types of variety combinations, the advantages of mixed plantation, and different aspects of levelling, lay-out and planting.

Chapter 12 contains advice and instructions on the shaping of trees and how this can be mechanized, and gives valuable directives as to thinning, rejuvenation and complementary pruning practices.

The achievements and experimental results obtained in recent years, which are presented in Chapter 13, under the title *Chemical Control of Growth and Yields*, deserve special attention. Numerous data, presented in column diagrams, figures and tables are used to illustrate the up-to-date production of propagation material (production of clones with synthetic auxins, modernized graft production using growth regulation of flowering and fruit set, and questions of fruit dropping and storability).

The subsequent chapters deal primarily with the practical aspects of plum cultivation and give a great deal of useful advice and instruction. They discuss the soil cultivation (14), fertilization (15), water regime and irrigation (16) and plant protection (17) in plum orchards, the harvesting (18) and processing (19) of plums, the question of mech-



anization (20), the profitability of plum production (21), and finally how to successfully grow, process and utilize plums in private gardens (22).

The book is completed by a rich bibliography, as mentioned above, and a well arranged list of contents.

It will be obvious, even from this brief review of the richly illustrated book, that the two authors, Dr. Elek Tóth and Dr. Dezső Surányi, and the experts who contributed to the individual chapters, have not only enriched agricultural and horticultural literature with a valuable, useful, much-needed work, but have also provided many useful results for use in scientific research.

S. SÁRKÁNY

J. SUTKA: *Cytogenetics*, Mezőgazdasági Kiadó, Budapest, 1980

The first Hungarian book that deals with cytogenetics on a firm scientific basis has been welcomed by professionals and the general reader alike, since we have finally been presented with a well-constructed, clearly formulated work discussing cytogenetics on an up-to-date level.

The book won a prize awarded by the Hungarian Ministry of Education in 1981 for one of the year's best publications. The award is a tribute to the author and the publishers alike. Besides its professional value, the high quality of the format, the clear structure and the abundance of illustrations are also praiseworthy.

The 243-page book includes 12 tables and 72 figures taken from books and publications written by well-known experts on genetics. These greatly facilitate the comprehension of the text. Of the 49 photos 47 have cytogenetic subjects. Most of the micrographs are originals, stemming from the author's own investigations or from related subjects (e.g. *Agropyron* addition). They are complemented by micrographs taken by Hungarian and foreign cytogeneticists.

The book is divided into eight chapters. The first gives a historical survey of the development of genetics, including cytogenetics. We are then made acquainted with microtechnical methods (preparation, chromosome banding, chromosome identification) and informed about the cytogenetic importance of tissue cultures. The morphology and structure of chromosomes, the cell cycle, and mitosis are discussed in separate subchapters.

In the second chapter the author discusses the phases of reductive cell division. This is followed by a clear description of

what is meant by synaptonemal complex, chiasma, crossing-over and recombination, and information is given on the genetic control of chromosome pairing (control of meiotic mutants, somatic association, homologous chromosome pairing). Finally, there are subchapters on gametogenesis and insemination in the fauna, and sporogenesis and double fertilization in angiospermous plants.

Chapter 3 offers detailed information about special chromosomes (polytene chromosomes, lampbrush chromosomes, sex chromosomes, and in this connection male and female heterogametic sex determination, B-chromosomes, ring chromosomes, telocentric and isochromosomes, di- and holocentric chromosomes). The unusually detailed summary of the occurrence, characteristics, hereditary role and phenotypic effect of B-chromosomes deserves special mention.

The chapter entitled "Structural changes in chromosomes" gives a characterization of the types of reorganization possible within the chromosome (deletion, duplication, inversion), the phenomenon of translocation and some aberrations caused by translocation, all well illustrated with examples and figures. A description of the effects of mutagens on chromosomes and hypotheses of the origin of chromosome aberrations are followed by the sub-chapter "Cytogenetic risks in our environment". In this section, which is of outstanding value and extremely important for a wide range of readers, the author presents data to underline the rapid increase in environmental pollution and takes a stand for the checking of this alarming process. It is to be hoped that the sometimes startling picture of the present situation will cause people to stop and think. The danger to humanity is clearly shown by the statistical data, according to which chromosome aberrations occur in some half (40–60%) of abortions taking place in the first three months of pregnancy, while about 5% of new-born children suffer from one or another hereditary disease. The diagram taken from Kihlman and Sturelid (page 117, Fig. 39), showing chromosome aberrations induced by eight mutagens when acting alone or in combination with coffee after-treatments, gives food for thought. The supplementary coffee treatment multiplied the original number of mutations in every case. The role of alcohol and nicotine in promoting similar chromosome aberrations is indubitable.

In connection with environmental pollution, modern cytogenetic tests suitable for indicating the chromosomal effects of mutagens are presented. These methods make it possible to screen for mutagens particularly dangerous to humans.



Chapter 5 discusses the major polyploid series found in the flora, the origin of polyploids, and the genetics of auto- and allopolyploids. This is followed by information on the genome state of some major genera. Under the heading "Haploidy" information is given about the origin of haploid plants and the possibility of producing them.

The chapter "Aneuploidy" supplies abundant knowledge, particularly on the genus *Triticum*. The discussion of trisomes is complemented by a separate section on trisomes discovered in humans (mongolism, Edward's disease, Patau's disease, Klinefelter's syndrome, XYY- and XXX-syndrome). The production of monosomic series in wheat, monosome analysis and chromosome mapping are the author's fields of research, so a number of original examples are found in this part of the book. Most of the cytological plates are connected with questions discussed in this chapter. The description of nullisomes and tetrasomes is followed by a clear, well-illustrated discussion of chromosome additions and substitutions, forming an integral part of the chapter.

Under the title "Evolutionary cytogenetics" (Chapter 7) information is presented on the chromosome variations in natural populations, the relationship between chromosomes and species formation, and certain possibilities of evolution in the flora. At the end of the chapter some idea is given of the phylogenetic relationship between Man and the primates.

In Chapter 8 the author discusses the connection between cytogenetics and plant breeding, showing ways in which cytogenetics can be utilized to produce new varieties. The results achieved with induced polyploids, triploid sugar-beet and triticale breeding are presented. One sub-chapter is devoted to the utilization of intervarietal and interspecific substitutions. The present achievements and future prospects of the practical application of haploid breeding, chromosome engineering plant protoplasts and somatic hybrids are discussed.

J. Sutka's book on cytogenetics is basically a summary of classical cytogenetics, of the knowledge acquired so far about chromosomes. In this respect it covers almost the entire scope of this branch of science. Some references to molecular genetics are also to be found, though fewer than could be desired. Despite the fact that molecular genetics has become an independent branch of science, requiring a separate text-book, a molecular genetic description of certain phenomena of cytogenetics would perhaps have further increased the value of the book.

We should have been glad to read somewhat more about the hereditary impor-

tance of other parts of the cell besides the chromosomes. Few experimental data are available in this connection, but the role of the cytoplasm in hybrid wheat production or in the instability of Triticale varieties is well known.

The references are correct, and can generally be found even for figures which are well known from earlier text-books. In a few cases, however, important sources are not indicated (e.g. lampbrush chromosomes of the oocytes of amphibians; statistical data on chromosome aberrations appearing in human populations in the text on page 123, or the origin of Agropyron additions shown in an original photo, Figs 38, 39).

It is a pity that the publishers (Mezőgazdasági Kiadó) did not publish the full bibliography. Those engaged in cytogenetic research will find the "Major literary sources" unsatisfactory, particularly when they require detailed information on some phenomenon.

J. Sutka gives short, clear explanations or definitions of the cytogenetic terms. An alphabetical list of these short explanations of the terminology would be a great help, especially for those not yet familiar with this field of science. The structure of the book renders it possible to look up the required technical term, but the majority of readers will probably not take the trouble to do so.

Owing to the author's field of research, most of the cytogenetic information given in the book refers to plants, though examples of human and animal genetics are encountered in many places, widening the scope of knowledge in a very creditable way.

Sutka gives an objective presentation of the sometimes complicated or even contradictory cytogenetic data. While this possibly makes orientation difficult for those unfamiliar with the subject, it offers a sound basis for the further investigation and evaluation of open questions. The discussion of the relationship between cytogenetics and plant breeding and of the possibilities of co-operation deserves special mention, particularly the unusually full presentation of Hungarian achievements. It is a pity that the language barrier will prevent this information from reaching more than a few people.

In spite of the limited number of people engaged in cytogenetics in Hungary, four thousand copies of the book were sold within a couple of months of publication. This shows that apart from those professionally involved in the subject (researchers, teachers) many people are interested in the secrets of the chromosomes. The book is recommended primarily to researchers and teachers of genetics. In addition, however, those interested in genetics with a view to its practical application may also obtain useful informa-



tion from it. Since it gives a short, clear summary of cytogenetic phenomena, the book may help to considerably deepen the professional knowledge of young people interested in genetics.

D. SZALAY

*Poljoprivredna Znanstvena Smotra. Agriculturae Conspectus Scientificus Universitatis Zagradiensis* (Scientific communications of the Faculty of Agronomy, University of Zagreb)

The 135-page publication bearing the title "Scientific Communications of the Faculty of Agronomy, University of Zagreb 50" encompasses a wide spectrum of this field of science, presenting the results of scientific investigations. The publication includes 9 original scientific papers, a preliminary report and a large quantity of professional information. The interest and participation shown by Croatia researchers in scientific public life is well-known to those working in international scientific organizations or attending scientific conferences, and the present publication gives further evidence of their high standard of research. Considering the present "endproduct-centred" attitude to agricultural production, the publication deals mostly with questions of livestock breeding and feeding. There is no doubt that up-to-date meat and milk production is at present at the focus of interest all over the world. Most of the co-authors of the publication discuss questions of breeding and feeding related with meat and milk production.

In the introductory communication S. Jaksic gives the reproduction parameters of the white porker and its crosses under large-scale keeping conditions, based on examinations of more than 25 thousand pig litters. The study presents the progeny results of many pure-bred stocks as well as of  $F_1$ ,  $F_2$  and  $R_1$  stocks; the data were collected over several years of breeding on the SLJEME pig farm. The large white, Swedish lowland and Dutch lowland breeds were included in the pure breeding and crossing. The large white  $\times$  Swedish lowland  $\times$  Swedish lowland  $R_1$  combination proved superior to the large white  $\times$  Swedish lowland  $F_1$  stock. Even better results were obtained when hybrid sows were mated with Dutch lowland boars. In the course of pure-bred crossing the Swedish lowland stock was found to be more productive than the large white or Dutch lowland animals.

Z. Crnojevic et al. carried out experiments to study, among other things, the effect of the date when piglets were weaned on the slaughtering and meat quality param-

eters of animals fattened to a final weight of 118–121 kg. The authors examined the relationship between mortality and the number of piglets in the litter at birth and at weaning. They also studied the meat yield, the composition of the meat, its water content, pH and iodine number as a function of the treatment.

D. Lovrinov studied the effect of calving age on the average daily milk production of cows calving for the first time. The cows examined were half or full siblings. The investigations were carried out on the VRANA farm. The author found that the age at calving did not significantly influence the milk production during the first lactation in the stock examined.

J. Lukac-Skelin describes examinations performed on milk cows, with the aim of determining the milk production value of lucerne silage, depending on whether the lucerne was ensiled with or without zinc-bacitracin. During the experimental period one group of animals was fed with lucerne silage by itself, while the others were given 5 or 20 ppm zinc-bacitracin added to the lucerne silage. The colour, smell, structure and chemical parameters of the silage, as well as the amount of milk produced during the feeding period definitely showed the superiority of the zinc-bacitracin treatment. The milk was of good quality, and no problems arose when producing fermented dairy products either.

On analysing the fatty acid and oil content of three different soya varieties R. Heneberg, M. Miric et al. found the oil content in newly produced varieties to be 22% compared to 18% in earlier varieties, which corresponds to the world standard. The climatic factors during the year of examination had a greater influence on the oil content of the soya than the genotypes of the varieties. Of the three varieties examined, Makszimirka had the lowest oil content under the given climatic conditions. In 1975 this variety contained a relatively small proportion (6.76%) of linolenic acid combined with more olein and linoleic acid than the variety Srecka, in which the linolenic acid content was 6.31%. The largest amount of oil was found in the variety Srecka in 1977.

When studying the effects of atrazine and alachlor I. Siljes found treatments with a combination of the two herbicides to be more effective than when atrazine was applied by itself. When a smaller proportion of atrazine was used in the herbicide combination the herbicide residue content of the oil decreased. The largest amount of herbicide residue was found in the uppermost 0–5 cm layer of the soil. Soil particles larger than 2 mm in diameter promoted the leaching of atrazine into



deeper layers better than soil types with finer structures. The humus content of the soil had a great effect on the adsorption of the herbicide. In soils containing up to 1% humus more herbicide residue was found than in the case of a 1–2% humus content. Higher soil acidity (pH values lower than 6) decreased the adsorption of the herbicide, and promoted the accumulation of herbicide residue in the soil pores.

J. Bedekovic gives an account of the crushing strength of maize grains when operating a separator at various rotation speeds. He establishes that a higher separator speed increases the proportion of broken grains compared to those obtained after manual or mechanical harvesting. Mechanically harvested maize always contains a larger number of broken grains than that harvested by hand. Great attention must therefore be paid to physical strength when breeding hybrid maize for grain, with a view to increasing the efficiency of mechanical harvesting, storage and preservation.

K. Dubravec discusses the application of growth regulators in vineyards. According to this author, if growth substances are used, less energy is required for pruning in vineyards intended for vine production, and mechanical harvesting is also facilitated. The growth stimulator ETHREL proved more effective for Italian Riesling than for Traminer. ETHREL also influenced defoliation in the varieties examined.

B. Stancel deals with some theoretical and practical questions of teaching agricultural economics. The author attaches great importance to a high standard of instruction in farm management and economic subjects.

#### I. HEROLD

*Trace element metabolism in man and animals.* Proceedings of the Fourth International Symposium on Trace Element Metabolism in Man and Animals (TEMA-4), held in Perth, Western Australia 11–15 May 1981. Edited by J. M. Gawthorne, J. McC. Howell, C. L. White. Springer-Verlag, Berlin—Heidelberg—New York, 1982

The interdisciplinary and international TEMA symposia survey the most recent 4–5-year results of microelement research and always offer some important discoveries too. The first meeting was organized in 1969 at Aberdeen, Scotland and its material was edited by C. F. Mills (Livingstone, Edinburgh and London, 1970). The next symposium was held in the United States, at Madison and the lectures were published by W. G. Hoekstra

et al. (Univ. Press, Baltimore, 1974). The third meeting took place in the West-German Munich, and the publications were prepared for the press by Kirchgessner et al., in 1977.

At the symposium organized in Perth, Western Australia, 210 scientists from 24 countries took part. This is a fairly large number if we take the increased travelling costs into consideration. Most of the participants — 88 in number — came from the host country. The United States was represented by 37, Great Britain by 22 and the GFR by 13 scientists. Besides them only New Zealand, Sweden and Japan sent 5 or more delegates. From the socialist countries no one was present, in contrast to the Aberdeen symposium which 17 representatives from the socialist countries attended.

The book contains the material of 147 lectures and a further 23 works published on posters. With a view to an early publication and reduced costs, an offset litho process and a "camera ready" system were used. The material of the debate had been recorded, typed, and during the meeting a possibility of correction was provided. Apart from some cases of retyping, no alterations were made in the manuscripts.

Of the participants' scope of interest, the best picture is given by the frequency of elements included in the titles of the lectures. Out of twenty-two elements, zinc was discussed in 41, copper in 40 and selenium in 34 cases; and besides them, only cadmium, iron, manganese and cobalt are found in the titles of five or more works. The other elements, e.g. arsenium and nickel, which filled a thick volume each at the symposium held in Jena, 1980, were the main subjects of two lectures each. It is also surprising that the interest in boron is so modest; although, decades ago, already convincing arguments could be read about its role in the animal organism.

In the introductory lecture C. F. Mills — contrary to practice — did not review the microelement research of the last years, but spoke in praise of Eric Underwood, an Australian researcher, who was prevented by his death in August 1980 from making the opening speech as president of the Local Organizing Committee. His investigations relating to cobalt, his books reckoned as standard sources of information, his role in the world of science both at home and abroad, all had much to do with the decision that the TEMA-4 should be organized in Perth, the place of his activity.

The lectures cover 13 fields; most of them — 23 in number — discuss microelement deficiencies, while analytical aspects are the subjects of 5 lectures only.

### *Trace Element Status and Requirements*

Five of the 16 lectures concern humans, 6 are concerned with cattle, but there also are observations on horses, deer and rats. In an interesting way, the sustenance requirements are largely the same if the daily requirement is related to 1 kg body weight. Thus, the zinc requirement is about 0.4 mg and the copper requirement 0.1 mg per kg body weight and day. The nutrition habits greatly influence the microelement intake. In Sweden, for example, an average adult consumes 8.5 mg zinc, while the vegetarians take in 11.8 mg zinc per capita. On the other hand, compared to the daily ration of 0.011 mg selenium for the vegetarians, the lacto-vegetarians consume 0.064 mg selenium a day.

### *Trace Element Balance Studies and Homeostasis*

Among the experiments of mostly human concern, I mention here the one in which was mentioned an increase in the amount of zinc excreted with urine and faeces, in response to a daily 50 mg intake of tin. At the same time, the zinc content of the serum did not change. On the basis of observations made by myself and others, I think that the extent of mineral supply can be measured on a much wider scale by the examination of excreta than by the analysis of blood. Any considerable change in the blood can only be detected in the case of a catastrophic breakdown of the controlling mechanisms.

### *The Availability, Absorption and Retention of Trace Elements*

Although partly known effects are also discussed (Zn—phytate—protein—tannin interaction, Cu—Mo—S interaction, effect of complex forming substances, etc.), the techniques of experimentation is now more circum-spect, the results more convincing, than earlier. Many details have been cleared concerning the factors that influence the microelement metabolism.

### *Trace Element Supplementation*

Only one of the 17 lectures discusses human experiment, with the conclusion that a daily intake of 1500 mg Ca did not influence the Cu and Zn concentrations in the blood of elderly women. Seven articles give account of the effect of selenium supplementation, and another two try to approach experimentally the optimum size of selenium grains in the pellets. Observations related to copper repeatedly suggest the negative role of excesses of molybdenum and sulphur.

Some authors compare oral and parenteral microelement supplementations that they generally considered alternative solutions. Beside microelement compositions resting long in the digestive tract, glass-like supplements implanted under the skin or in the muscle are also discussed. The evergreen subject of anaemia in piglets has been enriched with a new therapy. The evolvement of anaemia was prevented by 2000 ppm iron, given to the sow with the feed in the form of ferrosulphate. Namely, the piglets could take up a sufficient quantity of iron from their mother's faeces. Surprisingly enough, the agrochemical methods of supplementing microelements (fertilization, alteration of the chemical reaction of soil, etc.) are not mentioned in this chapter.

### *Trace Elements in Pregnancy and Lactation*

In this chapter every work touches upon the subject of zinc metabolism, even if not exclusively dealing with it. Pregnancy involves an additional zinc requirement of some 375 mg. A seldom-encountered series of summarizing data is published on the composition of milk in nine species, with regard to the distribution of the element in butterfat, casein and whey, as well as in the early and later phases of lactation. One paper reports that the milk of women with higher incomes contains more copper and zinc, in the first three months of lactation, than does that of women with lower incomes. The oestrogen — progestagen type contraceptives have been found to increase the Fe, Mn and Cu content of the plasma, while the mere progestagen-type ones do not cause essential changes.

### *Trace Elements and the Development of Organs and Tissues*

Seven of the 10 lectures discuss morphological changes observed in the brain, with special regard to copper and iodine. Of the development aberrations, the relation of lesions caused by salicylate and penicillamine to zinc and copper supply was examined. By the nature of the subject, the enzymatic, histological and clinical observations prevail in these works.

### *Trace Element Deficiencies*

The subject is treated in 23 papers on 88 pages. Seven articles deal with the role of zinc in the metabolism, the diagnostics of zinc deficiency, its relation to the metabolism of protein and particularly of collagen,



and finally with its effects on reproduction, hormones and dermatitis. Six papers discuss the problems of myopathies, reproduction, calcification and phagocytosis in connection with selenium deficiency. Copper deficiency is described as reflected in biochemical changes, enzyme activity and catecholamine synthesis. Two works on cobalt deficiency deal with the B<sub>12</sub> vitamin and the fat metabolism, as well as with the increase of wool and body weight. Iron deficiency is described in relation to muscle work, and chrome deficiency is shown with the diagnostic application of hair. Although all articles represent totally new aspects, the one of outstanding importance points out the essential nature of lead. Accordingly, lead deficiency causes retarded growth, hypochrome microcytic anaemia and disorders in the iron metabolism, which phenomena can be prevented by supplying lead. The relationship between boron and arthritis, and the supposed relation between boron and neoplasms deserve attention. Namely, in spite of the evidences given by Bussler and Kovalskij et al., of the importance of this element for all living organisms without exception, most authors acknowledge its role only in plant physiology.

#### *Trace Element Environmental Contamination and Toxicity*

Cadmium is the subject of 8 lectures. The method of determining low concentrations with an isotope dilution technique, the origin of cadmium contained in hair, the possibilities of intruterine uptake, the kinetics of uptake, accumulation in the kidney and the effect exerted on the development of renal stones are investigated. Four works dealing with copper discuss the tolerance, the change of biochemical parameters preceding the haemolysis, the changes of kidney functions, and the increase in the Se concentration of liver on copper intoxication. In lectures with lead as their subject, the lead content of air, the effects of various carbohydrates influencing the incorporation of lead, and a possible relation between mental capacity and lead-poisoning are the questions examined. In the last work the possible role of cadmium is also considered. As regards fluoride, the balance of intake and discharge, and the revision of F concentrations tolerable by cows are mentioned. Mention is also made of the relation of the working of the nervous system to arsenic, of the iodophorous compounds and the iodine content of milk, as well as of Se concentrations tolerable by swine. Finally, a publication commissioned by the National Academy of Sciences (USA), in which the tolerance of domestic animals to 36 elements was summarized, is reviewed.

#### *Trace Elements and Human Disease*

This series was introduced by a lecture surveying the role of copper and zinc, which prompted a hot debate. As regards zinc metabolism, acrodermatitis enteropathica is discussed in connection with babies fed on lactose-free milk, further on in the course of an alopecia therapy, and in relation to the etiology of the coeliac disease. Diphenylhydantoine, as a chelate-forming compound, influences the metabolism of zinc and copper. An account is given of copper compounds absorbed through the skin and inhibiting inflammations. The role of selenium is discussed in connection with cystic fibrosis, myocardial infarction, and the so-called Keshan disease, which is a selenium deficiency disease occurring in China, that can practically be eliminated with a 0.5–1.0 mg weekly dose of selenium given to children. However, only some 2 per cent of the agrochemically applied selenium appear in the grains of cereals. Fluorosis is described from areas in India where the drinking water contains 3.5–6.0 ppm fluor. The chrome content of the hair is analysed in the case of arteriosclerotic heart diseases, while bromide deficiency is studied in patients with chronic haemodialysis, suffering of insomnia. The blood of those suffering of kidney-, liver- and gall-bladder diseases is analysed for a number of elements; and the liver is examined in connection with the sudden-death syndrome among infants.

#### *Trace Element Interactions*

The papers either reveal further details of known phenomena or discuss new interactions. The chapter clearly points out that deficiency or excess of any single element cannot by itself be numerically characterized, since this is essentially influenced by other inorganic and organic matters. New light is thrown upon the correlations of Cu—Mo—S turnover in cattle, upon the role of sulphur anions, wolfram, and thiomolybdates in the metabolism of animals. Copper and molybdenum are found to have no essential influence on the selenium metabolism of sheep. Zinc influences the absorption or excretion of a number of substances, causes copper deficiency in swine, increases the Cd and decreases the Se incorporation in rats. The effect of zinc on the absorption of vitamin A and on the metabolism of lead, iron and copper is also described. An increase in the concentration of calcium decreases the incorporation of cadmium in swine. Higher rates of cadmium intake increase the Ca/Mg ratio of rats' teeth. Definite Ni—Fe—Cu interaction is observed in rats. Evidence given of the boron — chol-



calciferol interaction in chicks indicates an important step towards the acknowledgement of boron as an essential element.

### *Functions of Trace Elements*

The series partly analysing special details is introduced by a survey of the relations between the elements and the immune system. The number of literary citations — 98 — and the vivid discussion equally show the great theoretical and practical importance of the subject. Accounts are given of the zinc—tubulin correlation, of the role of alcohol in raising the Mn/Cu ratio of the liver, of the synthesis and decomposition of selenocystein, of vanadium in the haemoproteins, as well as of the characteristics of ferritin in the different tissues. Of some aspects of nickel metabolism, it is worth mentioning that the Ni requirement of sows — as determined in balance experiments — is 0.7 ppm.

### *Metallothionein*

These proteins are characterized by a high metal content, low molecular weight, high cystein content and the lack of aromatic amino acids. After the introductory lecture that surveyed about two decades of investigations, experiments are described that relate to the copper-containing metallothioneins and the effect of sulphur intake on the synthesis of these proteins.

### *Aspects of Trace Element Analysis*

The alpha and omega of the microelement research, the measuring technique, was not given a central place at the meeting. The introductory lecture that disclosed many sources of error, made in sampling biological substances, referred to 75 literary data. The lecture on the use of stable selenium isotopes, X-ray microanalysis and application of tissue cultures has analytical implications, and so has the one that revealed the causes of differences between plasma and serum.

The names and addresses of participants at the end of the publications will greatly facilitate correspondence. The list of names of authors and those others who took part in the debates, with page references, occupies 5 pages; and the detailed subject index, an additional 13 pages. This book of 715 + XV pages, with its 17×25 cm size, is easy to handle; and, due to its narrow margin, contains a vast store of information.

Of the microelements that have a great effect on the health and performance of humans and animals, the best survey is provided by the TEMA congresses. Through the medium of lectures published in the form

of a book, many researchers can attentively follow the most recent world-wide developments in this field of science. As well as a positive source of data, important information is given on those centres of the world where large collectives conduct regular microelement experiments. Studying the book makes it possible to avoid rediscovering already discovered rules. Namely, the references reveal that even those living on the same linguistic areas either do not know, or do not want to know, about important precedents.

The typography of the book makes it easily readable. The figures, pictures and tables, although from different sources, do not break the impression of unity. The cover of the book also seems sturdy enough to endure frequent use.

This book is indispensable for those who were not present at the congress, and also for those who want to recall details again and again. With its material equally containing data and ideas, practical knowledge, and research suggestions, the book is recommended to physicians, veterinarians, agriculturists, biochemists, and to all those engaged in environment protection and nutrition science.

GY. TÖLGYESI

*Proceeding of Symposium on Paddy Soil.*  
Institute of Soil Science Academia Sinica —  
Science Press, Beijing, Springer-Verlag, Berlin, Heidelberg, New York, 1981\*

The "Proceedings of the Symposium", prepared in 864 pages with 317 figures and 445 tables, is a particular and good summary of the meeting held in Nanking in 1980.

Rice, as is well known among world food supplies, is the most important cereal. About half of China's food production is rice. From 100 000 000 ha arable soil in China, 30 000 000 ha (30%) is producing rice.

The symposium organized by the Academia Sinica (Beijing) was held 19–24 October 1980, in Nanking, and was followed by a seven-day paddy soil excursion in the Changjiang Delta. Beside 120 Chinese specialists, 56 invited soil scientists attended the symposium from America, Asia, Europe and Oceania. Altogether, 110 papers were given at the symposium, or presented by poster. All these are collected in this volume.

The volume consists of three main parts; first, the plenary sessions in 231 pages;

\* Materials from the Symposium under the auspices of Academia Sinica held on October 19–24, 1980, Nanjing, China.

second, the section papers in 458 pages; third, the publication of the posters.

Beside the plenary sessions, the section meetings were held on the following subjects;

- I. Section: Properties of paddy soils.
- II. Section: Genesis and classification of paddy soils.
- III. Section: The use and management of paddy soils.

#### *Plenary session*

The introductory paper was given by N. C. Brady, Director-General (International Rice Research Institute Los Banos, Laguna, Philippines). Brady outlined in his paper that no other main food plant has a greater sphere in soil and climate conditions than rice. Rice production is carried out on all continents between the 40–53° parallels of latitude. In arid conditions, rice production is similar to that of wheat, with irrigation on partly flooded or continuously flooded surfaces. The picture also varies greatly according to altitudes of soil surfaces from sea level. Rice is produced on alluvial soils in deep meadows as well as on slopes of terraced hills.

Among the properties of soil, physical conditions are lesser characteristic. Despite that, the sticky, bounded soils are preferred — partly because of their easy irrigation without water losses — partly because of their higher productivity. The lower level of redox potential and the possibility of higher buffer-capacity is also favourable.

The history of Asia shows that continuous rice production, without decrease of the product, is possible on suitable soils by suitable methods.

Regarding the uptake of plant nutrients, the outstanding characteristic of rice is its great nitrogen uptake, connected with high selectivity. First of all the  $\text{NH}^+$  form of N is preferable. According to Japanese results, for 5–6 t/ha production it is necessary to use 120–150 kg of N fertilizer, for N supply.

At present, all the rice-producing countries can be divided into three main groups, according to the applied amount of N fertilizer:

- I. Japan, South-Korea, Egypt 149 kg/ha/N
- II. China, Iran, Indonesia,  
W-Malaysia 30 kg/ha/N
- III. India and other rice  
producers of Asia 13 kg/ha/N

Also, Indian research workers explained the fact of how great is the importance of fertilization in rice production.

The other main plant nutrient P, and its supply, is very important for rice production

in Asia, because the soils there contain this element in much lesser amounts than is needed by rice.

Rice production is favourably affected by the availability of P, which nutrient also has great importance in assisting the absorption of N.

The effect of K on rice production is not less than the effects of N and P. Experiences in Japan, as well as in China, show that the potassium requirement of rice would be increased at the same time that the average production of rice increases.

The new intensive species of rice, compared with the traditional, extract 4 times the amount of soil potassium. The species used at the present time require 135 kg/ha  $\text{K}_2\text{O}$  by 5–6 t/ha of product.

From the microelements, rice is most sensitive to Zn deficiency. Indian research workers have achieved good results by the application of Zn.

It was also stated that rice has a considerable S requirement, almost equal to its K requirement.

It is interesting to consider that Si is not listed among nutrition elements, although this element is also necessary for good rice production. The production of rice is influenced by toxic processes also. There can appear, for instance, a Fe-toxicity. The acidic conditions, that cause reduction, act first of all on the S-toxicity, which is an inhibiting factor because of its acidic sulphate effect. This phenomenon is characteristic in Thailand and Vietnam.

Chen Zia-Fang and Li Shi-Ye have found that a favourable ratio among nutrients is very important for paddy soils with high production. The optimum humus content is between 2–4%. Below 2% is it insufficient, and also unfavourable above 4% because of the dominance of anaerobic processes. The application of lesser amount of fertilizers preserves the humus content and its characteristic N amount. For production above 6 t/ha this is unimportant.

The suitable management of soils gives a good permeability to plant roots. The authors have found a positive correlation between the thickness of the cultivated layer and the root density in the same soil layer.

Dudal, Hrabovszky and Précot, representing the FAO, investigated in their paper the role of rice soils in world food production. Some countries have 6 t/ha average product, with the application of intensive species. On the other hand, the average product level for the whole world is only about 2 t/ha. Beside species considerations, the soil and irrigation conditions are most important. According to thousands of years experience in China, organic manuring has an outstand-



ing effect on the maintenance of good soil conditions.

Greenland explained the importance of soil structure. Humus plays an outstanding role in the development of this structure. Soils with low humus content are affecting through the polysaccharides on the agglomeration of particles considerably. In soils with higher humus content, specific substances in the humus act in the same way. Overflooded conditions are considered very important for root development.

Ponnanperumpa has given an outstandingly bright picture of physico-chemical properties of paddy soils. He outlined how the decrease of redox potential reduces the nitrates and increases the mobility of phosphates, as well as the mobility of Mo, by overflowing. Simultaneously, the availability of Cu and Zn decreases.

Fe and Mn act favourably upon the mobility of basic cations at first, but they increase acidity. Temperatures above 25 °C increase the N loss by  $\text{NO}_3$  decomposition. The adsorption of  $\text{Fe}^{++}$  ions is considerable on organic matter, and also of interest is the result that intensive decomposition of organic matter is bounded to the highest Fe adsorption. Also great is the adsorption of  $\text{Mn}^{++}$  ions on organic soil matter. The results of the characterization of redox conditions were set forth in the paper of Yu Tian-Ren, who explained that it is very important to continue research work to obtain more knowledge of both soil redox systems and the role of humic substances in the redox conditions.

Patrick investigated the properties of inorganic redox systems and described their role. In his paper he explained that iron compounds in their oxidized forms play a major part in the decomposition of organic matter in overflooded paddy soils.

The paper of Kyuma (Japan) dealt with the use of tropical soils in Asia for rice production, and with classification of paddy soils. The subject was outlined through different methods by computer programs.

Gong Zi-Tong (China) explained that the need for paddy soil classification was first recognized in China in the 1930's. For instance, the expression "W horizon" was first introduced for paddy soils in China in order to characterize a special sublayer of B-horizon, developed by illuviation. The paper also presented an overview of soil geography and classification of paddy soils in China.

The paper of Mormann summarized similar problems. According to this author, it is possible to include the various paddy soils in the frames of present taxonomy. In some special cases, it is very necessary to provide new taxonomic units in the present taxonomy, for instance by accumulation of Fe and Mn

together. Xiao Ze-Hong explained that the paddy soils with low fertility can be reclaimed with rational irrigation. The P fertilization may be specially effective on the soils with low production in China.

Several authors dealt with fertilization of paddy soils; Lu Ru-Kun, referring to world production, stated that the total world rice area is 150 mill. ha. The total product is 384.5 mill. t. The average is: 2.65 t/ha.

Area of paddy soils in China is 25.3 mill. ha, product 136.9 mill. t, average 3.98 t/ha.

Area of paddy soils in India is 40.2 mill. ha, product is 80.7 mill. t, average 2.01 t/ha.

Area of paddy soils in Japan is 2.5 mill. ha, product is 16.4 mill. t, average 6.42 t/ha.

The applied fertilizers in the above-mentioned countries were for N urea ammoniumsulphate ammoniumdicarbonate. Of these fertilizers, the last is lowest in efficiency.

The use of P fertilizers is not only important from the standpoint of P supply, but also for increasing the effect of N fertilizers. Greater doses of potassium fertilizers have a primary importance for a high rate of fertilization.

Datta, Stangel and Groswell analyzed the efficiency of N fertilization. N efficiency in overflooded areas is only 30–50% of the used N fertilizers. The reason may be the volatilization of ammonia, the denitrification and biological immobilization by algae. Ammonium adsorption and elution also plays a role. According to Datta, sulphur covered urea provides the possibility for better results at about 6.3 t/ha for the highest yield.

Gu Rang-Shen dealt with problems of green manuring. In China green manuring has played an important role since the third century. It is also the Chinese experience that the most favourable yield in biomass can be developed by green manuring, along with the best grade of humification of the organic materials.

Xu Qi mentioned that the experience of rice production during 5000 years in China also influences the level and yield today.

#### *The papers of the sessions*

##### *1. Session. Properties of paddy soils*

From the experience in the use of reclaimed alkali soils for rice production to the equilibrium of the soil sulphides and the factors influencing the redox potential, as well as the reductional effect of the soil organic matter, a wide range of subjects deal with particular analysis of the physico-chemical properties of paddy soils.

The analysis of chemical properties include, beside the investigations of properties of organo-mineral complexes, the effect of



liming on the availability of microelements. In connection with the investigations of chemical properties, the special problems of environment contamination were also investigated, as were the problems of  $^{90}\text{Sr}$  contamination and the effect of HCH residues. Concerning the latter, it was stated that the amount of residues is inconsiderable. Among the 4 isomers, the gamma isomer is found in the lowest level. That shows the importance of production and use of the gamma isomer.

The biochemistry of microbiological action in the soils is always very important in rice production and was investigated in its many relations. Of all 23 papers presented on the symposium dealing with properties of paddy soils, 39% referred to these last mentioned problems.

The N fixation transformation and reduction were the main questions on the subject of N supply for paddy soils. Also, special investigations characterized the N fixation by the azolla and anabaena species. These problems are traditions of investigation in China, because it has long been known that the N supply transported by algae is considerable.

### 2. Session. *Genesis and classification of paddy soils*

Half of the 20 papers given in this session dealt with genesis and classification; the other half, with properties and regional distribution of paddy soils, as well as with special questions concerning these soils.

All the questions of classification and genesis as well as the results of various paddy soils, mainly referred to paddy soil areas of China and Japan. The particular characterization of the soil types concerns the illuviation of soils, as well as their microbiological properties — with role of clay minerals and properties of iron compounds in the soils.

Some research works dealt with magnetic susceptibility of soils. Chinese research workers tried to characterize paddy soil regions through their magnetic susceptibility, measured in the soil layers.

### 3. Session. *Management of paddy soils*

23 papers concerned management, production technology and relationships of soil manuring, including the scientific problems of drainage and the use of waste waters.

Research work dealt with both macroelement and microelement supply.

Investigating the conditions of paddy soils, some researchers stated that the regeneration effect of agriculture in environmental protection is much more characteristic than the effect of environmental contamination. In connection with this, they also investigated the reusing of waste waters in rice production.

The papers dealt with the role of the rhizosphere in N supply as well as with the technology of fertilization. The effect of nitrogen fertilizers under cool climate conditions was investigated, according to experiments in Hokkaido, Japan, where it was shown that higher yield could be expected by organic manuring than by N fertilizing. For this reason, methods were outlined for slowly-acting top dressing. The loss of N by fertilization is also possibly decreased, especially when N fertilizers are added together with organic manures. Several papers also dealt with questions of N fertilization as well as with the loss of N.

A symposium paper also discussed potassium content of paddy soils in South-East Asia. For such soils, which have medium or high K content, it is valuable to measure the ability of the K supply in long-term experiments. The paddy soils and their K status in China were summarized in a paper on the work of the Experiment Station for Potassium, Büntelhof, GFR. Within this paper, the relationships between soil types, their clay minerals and potassium supply were investigated. One of the papers in this session proposed the acceptance of peroxidase activity for characterization of potassium status determined in the leaves of rice.

Comparisons of the productivity of paddy soil in various regions brought results that dealt with humus status and N content, as well as differences in the distribution of organic materials. It was concluded that the total content of organic soil matter, and the humic acid: fulvic acid ratio, decreased on the one hand from East to West, on the other hand increased according to distance from the sea.

### *Poster Session*

The proceedings also summarize the results of the poster session. Together with the systematic structure of the symposium and its proceedings, it was possible that we could have a description of macroelement supply, as well as the supply of microelements, in connection with their effect in the soil.

Some part of the papers dealt with sulphur problems of paddy soils in South-China. The authors explained that, in the hills of South-China, the sulphur content may decrease and S deficiency may appear. A method was developed according to which, by plantation of young rice plants in paddy soils with S deficiency, the plants are inserted in a suspension of gypsum or sulphur before planting.

According to content and distribution of S, the soils can be variously categorized. The



first category is characterized by the phenomenon of the absence of requirements for S fertilization. In the second category, there appears some S deficiency and the requirement for S fertilization. In the third category, S fertilization is continually needed.

Many results of research refer to the role of Zn, because rice is sensitive to Zn deficiency. By fertilizing the paddy soils, that show Zn deficiency with  $\text{ZnSO}_4$ , production could show, in extreme cases, 17% increase.

In connection with N fertilization, comparative experiments by models have shown that the losses by application of urea are greater than by ammoniumsulphate.

Some results pertain to Hg contamination of paddy soils. Colleagues from the Institute of Soil Science of the Academia Sinica, Nanking, classified three categories of Hg contamination in paddy soils. Contamination below 48 ppb Hg in the soil is practically no contamination. Paddy soil with 48–150 ppb Hg is slightly contaminated. Above 150 ppb, Hg contamination becomes very considerable.

Some results summarized investigations of the bacteria living in the rhizosphere of rice and their microbiological activity in N supply. It was concluded that the free-living N-fixing bacteria are, in their numbers and effects, much greater when close to the roots of rice. Thus, it is very important to investigate this phenomenon in the future.

Many results referred to the special properties of paddy soils. For instance, in the zone of red soil are found greater amounts of metallic elements.

Of high interest is the question of decreasing fertility in paddy soils. Double and triple cropping systems of rice do not lead to any decrease in humus content during 5 years by 2.5% original humus content. The authors dealing with such questions propose a triple cropping system, with a rotation rice-rice-feed crop, the highest yield to be obtained by rice-rice-rape rotation.

The high fertility of soils in Shanghai and its suburbs reflect their favourable water and drainage conditions. The cultivation and preparation of soils before rice planting is of much greater importance than that for other crops.

The last chapter of the book is very interesting, as it concerns symptoms of deficiency and problems of nutrient supply. Among these it mentions the deviation from suitable N : K ratio which leads to chlorosis. Potassium deficiency appears in the bronzene leaves. The elimination of S deficiency is very successful in South-China by the application of S fertilizers. Too much N in the paddy causes relative phosphor and potassium deficiency. Too much Ca content in the soil causes Zn deficiency in rice.

The mineralization of N in paddy soils is also interesting from the point of view of priming effect. Application of urea prevents the phenomenon of priming effect from appearing.

Immediate correlation between available P content of soils and grain production by rice could not be stated, but P was found favourable in acting upon other crops in the rotation.

Investigations of the potassium effect have shown that this effect could be investigated only by consideration of all the elements that enter into the activity of rotation.

Some papers also presented the effects of Fe, Cr, Zn, Mo. There were data in experiments that still left open any conclusions about availability of Fe and Cr. Both elements are outstandingly accumulated in the roots. The accumulation of Fe is 1/100 part and of Cr 1/10 000 part in all the other parts of the plant, compared with the concentration in the roots. Iron has a greater mobility in soils with weak aeration, as similarly is the mobility of Cr.

It could be concluded that the lowest B content is found in very calcareous or in red clay soils. Soils improved on granitic rock material or riolites have also a very low B content. B deficiency generally appears in acidic soils. The effect of Mo on rice could not be immediately shown, but this element acts primarily on the other crops in the rotation.

Of extreme interest is the result of Japanese research work, that soil productivity and its relation to rice production is affected not only by N fertilization but also by the organic matter content of soil.

The book closes with three general chapters; Sombroek — the General Secretaire of the ISSS — summarized the relation of Chinese science to the international soil sciences.

The experience of Chinese science in the subject of soil conservation is very precious, as also are Chinese results in their agricultural policy concerning the use of soils. The role taken by China, within the international network of model experiments on soil use, is also very important.

Bentley stressed in his paper "Soils and people" that the importance of soils must be considered in the future by development of food production worldwide, and to eliminate problem zones with standing food deficiencies. This also lies in the responsibility of the state offices and governments. The paper appreciated the efforts of China in this line.

Thorp American professor in soil sciences stressed the thousands of years of tradition in China on the use of soils. Chinese soil



science made contact with the soil science of Western countries in the 1930's. Since then, there began the preparation of soil mapping in the geological institutes. In the 1950's Thorp also edited a summary of the soil geography of China. The paper praises the symposium as an important step in the development of Chinese soil science.

This volume sums up the results of the Symposium on 864 pages, with particular information and with a well-fitted compilation of the whole wide field of the very important subject of paddy soils.

L. HARGITAI

*Plant Carbohydrates II. Extracellular Carbohydrates* (W. TANNER and F. A. LOEWUS eds). Encyclopedia of Plant Physiology. New Series Vol. 13B. pp. 1-769. Springer-Verlag, Berlin, Heidelberg, New York, 1982

Much encyclopedic knowledge has accumulated on the biochemical-physiological aspects of plant carbohydrates. Therefore, the carbohydrates are presented in the "New Series" in two volumes. The 2nd volume on this topic deals, somewhat arbitrarily, with the "extracellular" carbohydrates, meaning those carbohydrates located outside the plasmamembrane. Many of these compounds have important biological functions, such as recognition phenomena in host-parasite relationships or pollen-pistil interactions, symbiosis and secretion (export of carbohydrates). More "conservative" areas, like the synthesis and structure of cell walls (or substances on cell surfaces in general) have, however, also developed considerably.

The vast material, written by 37 authors, is divided into five sections: I. Cell walls of higher plants. II. Cell walls of algae and fungi. III. Export of carbohydrate material. IV. Cell surface phenomena. V. Lectin-carbohydrate interactions.

I. A high number of papers deal with the cell wall. "The constitution of plant cell wall polysaccharides" written by G. D. Aspinall, is an introductory chapter on the classification and main structural features of cell wall building complex carbohydrates. The general aspects of the "Ultrastructure of the plant cell wall" is discussed from a biophysical viewpoint by J. R. Colvin. A short note on "The assembly of polysaccharid fibrils" is written by D. G. Robinson. A counterpart of this biophysical viewpoint is a summary of the biochemical viewpoint of the "Ultrastructure of the plant cell wall", and is summarized by K. Kato. Logically, the following chapter discusses the "Biosynthesis and metabolism of cellulose and noncellulosic

cell wall glucans", including the involvement of lipid-intermediates and the possible role of hormonal control of cellulose biosynthesis, written by G. Franz and U. Heiniger. The next, very detailed chapter is by G. B. Fincham and B. A. Stone on the "Metabolism of noncellulosic polysaccharides", including ontogenetic changes and environmental responses. Special attention is devoted to the "Glycoproteins and enzymes of the cell wall", including peroxidases and glycosidases by D. T. A. Lampion and J. W. Catt. The fashionable subject, "The role of lipid-linked saccharides in the biosynthesis of complex carbohydrates" is superbly reviewed by A. D. Elbein. T. Higuchi summarizes the much-studied but still not fully understood field of "Biosynthesis of lignins". Although much of the details remain hypothetical, some of the regulatory aspects are already emerging. P. E. Kollattukudy, K. E. Espelie and C. L. Soliday present a review of the "Hydrophobic layers attached to cell walls. Cutin, suberin and associated waxes". Finally, the ever-green problem of "Wall extensibility: hormones and wall extension" is discussed by R. E. Cleland.

II. Structural aspects and biosynthetic considerations dominate the review by E. Percival and R. H. McDovell: "Algal walls — composition and biosynthesis". Reasonably enough, further related chapters follow, such as "Algal walls — cytology and formation" by D. G. Robinson and "Algal wall-degrading enzymes — autolysins" by U. G. Schlösser. An introductory chapter by J. G. H. Wessels and J. H. Sietma provides general information on "The fungal cell walls". Then, detailed attention is paid to various individual compounds playing an important role in microorganisms: "Chitin: structure metabolism, and regulation of biosynthesis" by E. Cabib; "Fungal glucans — structure and metabolism" by G. H. Fleet and H. J. Phaff; "Mannoproteins: structure" by R. E. Cohen and C. E. Ballou; "Biosynthesis of mannoproteins in fungi" by L. Lehle.

III. The problem of export of carbohydrate material is nowadays much in the center of interest. Again, the special problems encountered in fungi and higher plants are separately dealt with. "Secretory processes — general considerations and secretion in fungi", an introductory chapter, is written by R. Sentandreu, G. Larriba and M. V. Elorza and is followed by a review of J. H. M. Willison on the "Secretion of cell wall material in higher plants", and by a paper of M. Rougier on the "Secretory activity of the root cap".

IV. The section on cell surface phenomena includes two highly up-to-date, fascinating reviews, such as "Defined components in-



volved in pollination" by A. E. Clarke and "Carbohydrates in plant-pathogen interactions" by T. Kosuge.

V. Lectin-carbohydrate interactions are discussed from two points of view: "Lectins and their physiological role in slime molds and in higher plants" by H. Kauss and "The role of lectins in symbiotic plant-microbe interactions" by E. L. Schmidt and B. B. Bohlool.

Volume II of "Plant Carbohydrates" nicely completes the first volume. The standard is the same both as far as the choice of

competent authors and the level of their work is concerned. It is perhaps, worth stressing that microbiology and plant physiology (*sensu stricto*) overlap more strongly in this volume than in many others of the Encyclopedia. The same would be a mistake in many fields (e.g. nucleic acids and proteins) but in this particular area, I regard it to be an advantage. It certainly adds to the interest in this book, for a number of workers engaged in applied, microbiological research.

G. L. FARKAS

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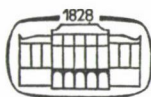
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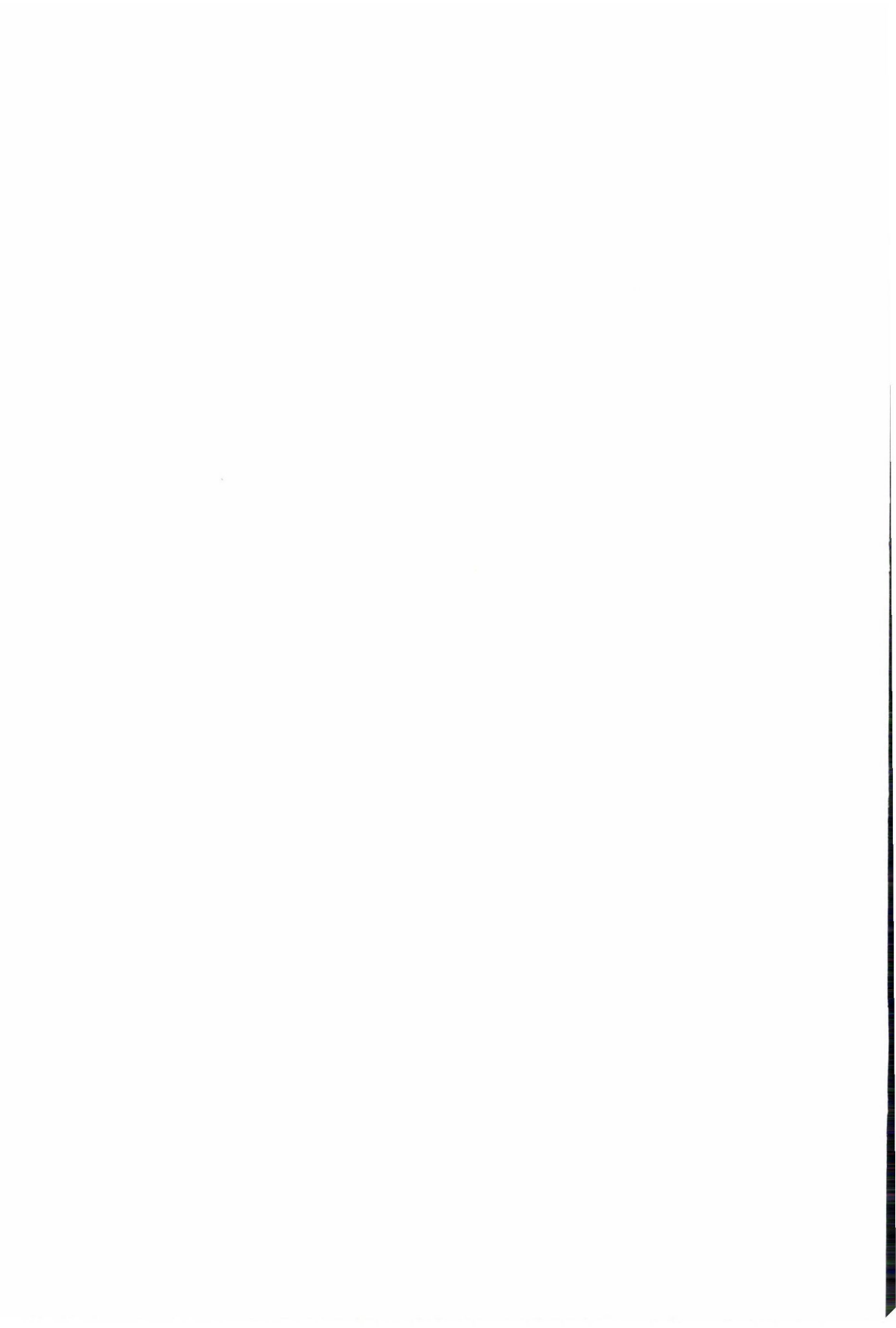
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## CHANGE OF SEX EXPRESSION OF SOUR CHERRY VARIETIES BY ROOTSTOCKS

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Between 1976 and 1980, the author studied the sour cherry varieties "Cigánymeggy" C. 404, "Érdi nagygyümölcsű", "Meteor korai" and "Pándy" C. 101 on wild cherry. *Prunus mahaleb* and "Cigánymeggy" stocks, and the variety "Újfehértói fürtös" on wild cherry and *Prunus mahaleb* stocks. In the length of pistil and absolute and relative number of stamina, significant differences depending on the rootstock were found between the combinations. The most striking effect of rootstock was shown in the combination "Meteor korai"/*Prunus mahaleb* C. 2740; where, in comparison to the combination of the same variety with wild cherry and "Cigánymeggy" stocks, the pistil was longer by 6.1 and 13.8 per cent, the stamina fewer by 17.1 and 29.9 per cent, and the relative number of stamina lower by 20.7 and 45.3 per cent respectively. The Chi<sup>2</sup>-test and the *r* values proved a close negative correlation between the length of pistil and number of stamina; furthermore, the author noticed that, in the self-fertile varieties "Meteor korai" and "Újfehértói fürtös", an increase in the relative number of stamina was accompanied by decreased productivity; while, in the case of self-sterile varieties, it was the other way round. Years of studies on flower organization in sour cherry varieties, demonstration of rootstock effects and seasonal changes have supplied sufficient morphological information for the evaluation of the effects of recent chemical control experiments on flower organization.

### Introduction

The time of flowering in sour cherry depends on the beginning of fruit bud formation, the mineral nutrient status of the tree, the period of dormancy (MOLNÁR 1963), the environmental factors (RADULESCU, cit. BRÓZIK and NYÉKI 1980) and the rootstock (NYUJTÓ 1971).

In consequence of flower induction, the flower organs start differentiating, and in the last phase of differentiation the morphological character of pistil, stamina, sepals and petals is established. Flower organization takes place according to strict rules; the correlation between pistil and stamina is of special importance, as it has an impact on the fertility of flowers (SURÁNYI 1976, 1977, 1980).

In sour cherry, the macrosporogenesis is often irregular, resulting in an underdeveloped pistil (PODDUBNAJA-ARNOLDI 1964); the extent of morphological (and physiological) sterility of pistil is a genetically fixed property (OLDÉN 1965). The undeveloped gynoecium is manifested in a high responsiveness of flower bud primordia to climate (PHILP 1933), nutrients and

hormones (PODDUBNAJA-ARNOLDI 1964), frost before flowering (FIELD 1942), as well as in pathological susceptibility (LEMOINE 1970).

The proportion of fertile pistils may vary by the variety (NYÉKI 1974), clone (SCHANDERL 1934), individual plant (SCHANDERL 1932) and year (NYÉKI 1975). Three types of pistil were described for Spanish sour cherry by SCHANDERL (1932), two types for Schattenmorelle by RUDLOFF and SCHANDERL (1950) and for Montmorency by PHILP (1933). NYÉKI (1974) and NYÉKI and TÓTH (1975, 1976) also distinguished several types — exactly 5 forms — of pistil in "Pándy" and "Cigánymeggy" clones, of which type 4 and 5 give the highest percentage of fruit setting. In the course of examining 18 "Pándy" clones, NYÉKI and TÓTH (1976) found wide fluctuations in the shape and size of pistil; 35 per cent in the length of pistil, 26 per cent in that of the style, 25 per cent in the length of ovary and 40 per cent in its width.

As a consequence of progeny sterility and chromosomal imbalance, microsporogenesis may also suffer disturbance, as concluded from the results of FEJKIC (1970). Similar experiences were reported earlier by MALIGA (1954), BARG (1958), MURAWSKI and ENDLICH (1962). The remarkably high fluctuations in the number of stamens and ability of pollen tube formation of some varieties in certain years can also be explained by this. According to several years observations made by SURÁNYI (1969–1972), in some "Pándy" clones the pollen tube formation shows a 287 per cent fluctuation, and the number of stamens in certain "Cigánymeggy" clones a 169 per cent variation, although the sour cherry (and cherry) are known as species with stable stamen numbers (MORRISON 1964).

UBACSINA *et al.* (cit. BRÓZIK and NYÉKI 1980) have pointed out that in sour cherry the percentage of pollen germination varies with the ability of self-pollination; the pollen tube of self-fertile varieties reaches the micropyle sooner than that of self-sterile ones. According to the opinion of BRÓZIK and NYÉKI (1980), the intensity of tube growth is suitable to characterize the compatibility of a variety.

In our investigations on the morphology of the flowers, we found the pistil length to range from 9.55 to 14.27 mm and the stamen number from 31.75 to 34.75 on the basis of four years observations of 9 sour cherry varieties. In the partially self-fertile varieties, the relative stamen number generally shows a lower value (Montreule, Nagy Gobet, "Császár") than in the self-sterile sour cherries (Eugenia császárnő, Ostheimi, Nagy angol, Hortenzia királynő, "Cigánymeggy" C. 215 and "Pándy" C. 116). The pistil length and stamen number in the partially self-fertile varieties are better levelled by year and variety than in the self-sterile varieties (SURÁNYI 1977).

In the present study, flower organization in the self-fertile variety "Meteor korai" and "Újfehértói fürtös", the self-sterile "Cigánymeggy" C. 404, "Érdi nagygyümölcsű" and "Pándy" C. 101 grafted on wild cherry, *Prunus*



*mahaleb* and "Cigánymeggy" rootstocks is discussed. We wanted to disclose partly the flower organization of the new varieties, and partly the new aspects of rootstock effect, in order to work out the proper use of rootstocks.

### Material and method

Between 1976 and 1980, the varieties "Cigánymeggy" C. 404, "Érdi nagygyümölcsű", "Meteor korai" and "Pándy" C. 101 were examined in three combinations of rootstock, and the variety "Újfehértói fürtös" on two stocks. At the time of full blossoming, flowers from the middle of the bunch were collected, pistil lengths measured (PL) and stamina counted (SN) in 30 flowers per combination, and the relative number of stamina, i.e. the number of stamina per unit length of pistil (SN/PL) was determined.

In a plantation established in 1972/1974 we studied the flower organization of trees standing on the following selected rootstocks:

- on wild cherry C. 2493 the varieties "Érdi nagygyümölcsű", "Meteor korai", "Pándy" C. 101 and "Cigánymeggy" C. 404;
- on wild cherry C. 1132 the variety "Újfehértói fürtös";
- on *Prunus mahaleb* C. 2740 the varieties "Cigánymeggy" C. 404, "Érdi nagygyümölcsű", "Meteor korai" and "Pándy" C. 101;
- on *Prunus mahaleb* C. 2737, "Cigánymeggy" C. 404 and "Újfehértói fürtös";
- on "Cigánymeggy" C. 215, "Cigánymeggy" C. 404, "Érdi nagygyümölcsű", "Meteor korai" and "Pándy" C. 101.

The time of sampling varied between 9 April and 8 May, depending on the time of flowering; "Meteor korai" on "Cigánymeggy" C. 215 rootstock showed 2-6 days of delay compared with the wild cherry- and *Prunus mahaleb* combinations.

The stock combinations of the different varieties were examined as a function of the fruiting year, and compared to each other with a year and as a function of years on the basis of pistil length, stamen number and relative number of stamina.

The varieties (with stocks as replications) and the rootstocks (with varieties as replications) were also evaluated by variance analysis.

Since the changes of pistil length and stamen number were regular, they were analysed first by  $\chi^2$ -test; then, to determine the tendency of the correlations found, regression analysis was used.

On the basis of pistil length and stamen numbers of different frequency categories, we examined the possibility of correlations from the point of view of varieties and rootstocks; and, in the relation of variety and rootstock, tried to establish the characteristics of limit values of variety and stock dependent pistil length and stamen number.

In the course of regression analysis, the value of  $r$  was determined for every year and each combination; this meant a total of 70 calculations ( $FG = 28$  in every case). Totalled correlation analyses by combination ( $FG = 148$ ), variety ( $FG = 448$ ) and rootstock ( $FG = 748$ ) were also carried out, and finally the  $r$ -value of the linear correlation was determined on the basis of the total number of flowers ( $FG = 2098$ ) too.

### Results

The length of pistil in the varieties "Pándy" C. 101 and "Újfehértói fürtös" shows a slight change as a function of rootstock. The pistils of trees on *Prunus mahaleb* stocks generally are longer than those of trees with wild cherry- and "Cigánymeggy" stocks. Of the varieties, "Érdi nagygyümölcsű" and "Meteor korai" are characterized by the longest pistils, while "Cigánymeggy" C. 404 and "Pándy" C. 101 have the shortest pistils. The crop year dependence of the pistil length was very low, except perhaps the difference in 1978 compared to the other four years (Table 1).



**Table 1**  
*Changes in the pistil size of sour cherry varieties*  
 (1976–1980)

Variety	Pistil length, mm					Mean	SD5%	
	Rootstock	1976	1977	1978	1979			1980
<i>Cigánymeggy C. 404</i>								
wild cherry C. 2493		10.9	10.7	10.3	10.6	11.2	10.8	0.59
<i>Prunus mahaleb</i> C. 2737		11.5	11.3	10.9	11.8	11.5	11.5	
Cigánymeggy C. 215		10.8	11.0	10.2	11.5	11.4	11.0	
Average		10.9	11.0	10.5	11.4	11.4		1.39
<i>Érdi nagygyümölcsű</i>								
wild cherry C. 2493		14.6	14.7	14.0	14.9	14.0	14.4	0.38
<i>Prunus mahaleb</i> C. 2740		14.3	14.5	13.6	14.5	14.3	14.2	
Cigánymeggy C. 215		14.7	14.9	14.1	15.3	14.4	14.7	
Average		14.5	14.7	13.9	14.9	14.1		1.18
<i>Meteor korai</i>								
wild cherry C. 2493		13.6	12.9	13.3	13.3	12.7	13.2	1.21
<i>Prunus mahaleb</i> C. 2740		14.5	13.1	13.7	14.2	14.9	14.0	
Cigánymeggy C. 215		11.5	12.4	12.2	12.0	13.4	12.3	
Average		13.2	12.9	13.1	13.2	13.6		1.75
<i>Pándy C. 101</i>								
wild cherry C. 2493		11.3	11.3	10.5	11.2	10.6	11.1	0.47
<i>Prunus mahaleb</i> C. 2470		11.4	11.4	11.3	11.2	10.9	11.0	
Cigánymeggy C. 215		10.7	10.5	10.9	10.6	11.1	10.8	
Average		11.0	11.1	10.9	11.0	10.6		1.41
<i>Újfehértói fürtös</i>								
wild cherry C. 1132		12.7	12.1	11.8	12.0	12.1	12.2	1.16
<i>Prunus mahaleb</i> C. 2737		13.1	12.4	12.0	12.3	12.3	12.4	
Average		12.9	12.2	11.9	12.2	12.2		
<i>Varieties</i>								
Cigánymeggy C. 404							11.1	0.79
Érdi nagygyümölcsű							14.1	
Meteor korai							13.2	
Pándy C. 101							11.1	
Újfehértói fürtös							12.3	
<i>Rootstocks</i>								
wild cherry							12.4	0.36
<i>Prunus mahaleb</i>							12.7	
Cigánymeggy							12.2	

**Table 2**  
*Changes in the stamen number of sour cherry varieties*  
 (1976–1980)

Variety		Stamen number, n					Mean	SD5%
	Rootstock	1976	1977	1978	1979	1980		
Cigánymeggy C. 404								
wild cherry C. 2493		32.4	32.6	32.8	33.0	33.9	32.9	0.71
Prunus mahaleb C. 2737		32.0	32.3	32.4	32.8	33.3	32.6	
Cigánymeggy C. 215		32.9	32.7	32.7	33.2	34.8	33.3	
Average		32.5	32.6	32.5	33.0	33.9		1.26
Érdi nagygyümölcsű								
wild cherry C. 2493		39.6	39.6	38.4	40.2	39.5	39.5	0.96
Prunus mahaleb C. 2740		40.9	39.4	39.2	40.3	39.7	39.9	
Cigánymeggy C. 215		41.0	38.1	39.3	39.5	38.3	39.2	
Average		40.5	39.0	39.1	40.0	39.2		1.16
Meteor korai								
wild cherry C. 2493		36.3	37.7	35.4	33.8	31.1	34.9	2.91
Prunus mahaleb C. 2740		31.4	29.5	29.4	28.6	30.1	29.8	
Cigánymeggy C. 215		37.7	38.4	37.9	38.7	41.0	38.7	
Average		35.1	35.2	34.2	33.7	34.1		3.29
Pándy C. 101								
wild cherry C. 2493		31.1	31.5	31.7	32.0	32.0	31.7	0.68
Prunus mahaleb C. 2740		31.9	32.7	32.6	32.4	32.9	32.5	
Cigánymeggy C. 215		32.2	32.7	31.7	31.5	31.1	31.8	
Average		31.7	32.1	32.0	32.0	32.0		0.91
Újfehértói fűrtös								
wild cherry C. 1132		34.7	32.4	33.4	32.4	33.3	33.2	0.56
Prunus mahaleb C. 2737		34.1	32.0	33.1	31.2	32.2	32.5	
Average		34.5	32.2	33.3	31.8	32.9		
Varieties								
Cigánymeggy C. 404							33.1	1.17
Érdi nagygyümölcsű							39.2	
Meteor korai							38.7	
Pándy C. 101							31.7	
Újfehértói fűrtös							32.8	
Rootstocks								
wild cherry							34.4	2.13
Prunus mahaleb							33.5	
Cigánymeggy							35.8	

Table 3

Changes in the relative stamen number of sour cherry varieties  
(1976–1980)

Variety	Rootstock	Relative stamen number, n/mm					Mean	SD5%
		1976	1977	1978	1979	1980		
<i>Cigánymeggy C. 404</i>								
wild cherry C. 2493		2.97	3.05	3.18	3.11	3.02	3.11	0.16
<i>Prunus mahaleb</i> C. 2737		2.78	2.86	2.98	2.79	2.90	2.86	
Cigánymeggy C. 215		3.05	2.98	3.21	2.88	3.06	3.04	
Average		2.95	3.00	3.14	2.90	2.94		0.26
<i>Érdi nagygyümölcsű</i>								
wild cherry C. 2493		2.71	2.61	2.78	2.78	2.83	2.74	0.09
<i>Prunus mahaleb</i> C. 2740		2.87	2.72	2.74	2.80	2.78	2.78	
Cigánymeggy C. 215		2.79	2.56	2.77	2.57	2.73	2.68	
Average		2.79	2.66	2.76	2.69	2.78		0.10
<i>Meteor korai</i>								
wild cherry C. 2493		2.71	2.45	2.66	2.54	2.45	2.56	0.10
<i>Prunus mahaleb</i> C. 2740		2.16	2.26	2.13	2.01	2.03	2.12	
Cigánymeggy C. 215		3.23	2.87	3.11	3.23	2.95	3.08	
Average		2.70	2.53	2.63	2.59	2.48		0.16
<i>Pándy C. 101</i>								
wild cherry C. 2493		2.76	2.78	3.01	2.86	3.01	2.88	0.13
<i>Prunus mahaleb</i> C. 2740		2.79	3.14	2.89	2.90	3.22	2.99	
Cigánymeggy C. 215		3.02	3.13	2.90	2.96	2.80	2.96	
Average		2.86	3.02	2.93	2.91	3.01	3.00	0.25
<i>Újfehértói fürtös</i>								
wild cherry C. 1132		2.73	2.71	2.74	2.70	2.75	2.74	0.27
<i>Prunus mahaleb</i> C. 2737		2.60	2.58	2.76	2.54	2.62	2.62	
Average		2.67	2.64	2.75	2.63	2.68		
<i>Varieties</i>								
Cigánymeggy C. 404							2.91	0.19
Érdi nagygyümölcsű							2.73	
Meteor korai							2.59	
Pándy C. 101							3.13	
Újfehértói fürtös							2.68	
<i>Rootstocks</i>								
wild cherry							2.83	0.25
<i>Prunus mahaleb</i>							2.67	
Cigánymeggy							2.94	



Changes in the number of stamina are seen in Table 2; the stamen number as a function of crop year is very stable, except in the variety "Újfehértói fürtös". The effect of rootstock can also be pointed out in the flowers of the varieties, though the differences usually are rather small. The effect of *Prunus mahaleb* C. 2740 on "Meteor korai" trees is, however, remarkable; namely, the number of stamina in the outer circle is lower by about 10 than in trees standing on the other two rootstocks. As a whole, the stamen number in the varieties "Érdi nagygyümölcsű" and "Meteor korai" is essentially higher than in the varieties "Cigánymeggy" C. 404, "Pándy" C. 101 and "Újfehértói fürtös". The "Cigánymeggy" rootstocks mostly result in the formation of flowers with reduced numbers of stamina, though the lowest stamen number is found in trees grafted on *Prunus mahaleb*, compared to the other two rootstocks (Table 2).

The relative number of stamina depends on the rootstock, variety and to a minor extent on the crop year. The number of stamina per unit pistil length is the smallest in "Cigánymeggy" C. 404, "Meteor korai" and "Újfehértói fürtös" trees grafted on *Prunus mahaleb* rootstocks; low relative stamen numbers are shown by the "Érdi nagygyümölcsű" on wild cherry- and the "Pándy" C. 101 on "Cigánymeggy" C. 215 stocks (Table 3). The relationship of this effect of rootstock with fertility and productivity will be analysed later (cf. Fig. 1).

Earlier investigations pointed out a close correlation between pistil length and stamen number. A summation of data on the frequency groups of pistil length and stamen number, from the point of view of varieties and rootstocks, suggests the existence of a close relationship (Table 4). About the

Table 4  
Major results of  $\chi^2$ -test calculations

Variety Rootstock	Pistil length to stamen number (4 × 5) FG = 12	Variety or rootstock to	
		pistil length (3 × 4) FG = 6	stamen number (3 × 5) FG = 8
Cigánymeggy C. 404	17.12		
Érdi nagygyümölcsű	21.94		
Meteor korai	53.46	891.50	1021.83
Pándy C. 101	35.03		
Újfehértói fürtös	117.53		
Wild cherry	218.15		
<i>Prunus mahaleb</i>	179.55	23.42	96.06
Cigánymeggy	368.04		
Altogether	444.38		

**Table 5**  
Results of regression analyses

Variety	<i>r</i> -value of correlation					Together
Rootstock	1976	1977	1978	1979	1980	
<i>Cigánymeggy C. 404</i>						
wild cherry C. 2493	−0.37	−0.16	+0.28	−0.39	−0.51	−0.39
<i>Prunus mahaleb</i> C. 2737	−0.79	−0.86	−0.53	−0.47	−0.59	−0.88
Cigánymeggy C. 215	−0.52	+0.18	−0.35	−0.89	−0.53	−0.25
<i>Érdi nagygyümölcsű</i>						
wild cherry C. 2493	−0.36	+0.14	−0.13	−0.23	−0.78	−0.25
<i>Prunus mahaleb</i> C. 2740	−0.40	−0.29	−0.57	−0.09	−0.81	−0.17
Cigánymeggy C. 215	+0.01	−0.24	−0.14	+0.04	−0.41	−0.26
<i>Meteor korai</i>						
wild cherry C. 2493	−0.66	−0.61	−0.15	+0.18	−0.52	−0.25
<i>Prunus mahaleb</i> C. 2740	−0.01	+0.14	−0.26	−0.10	−0.73	−0.28
Cigánymeggy C. 215	−0.32	−0.18	−0.96	−0.83	−0.20	−0.16
<i>Pándy C. 101</i>						
wild cherry C. 2493	−0.56	−0.17	−0.16	+0.09	−0.86	−0.19
<i>Prunus mahaleb</i> C. 2740	−0.13	−0.73	−0.44	−0.26	−0.82	−0.21
Cigánymeggy C. 215	+0.05	−0.86	+0.13	−0.19	−0.54	−0.15
<i>Újfehértói fürtös</i>						
wild cherry C. 1132	−0.46	−0.31	−0.81	−0.33	+0.12	−0.40
<i>Prunus mahaleb</i> C. 2737	−0.90	−0.77	−0.29	−0.16	−0.69	−0.52
<i>Varieties</i>						
Cigánymeggy C. 404						−0.34
Érdi nagygyümölcsű						−0.19
Meteor korai						−0.20
Pándy C. 101						−0.36
Újfehértói fürtös						−0.51
<i>Rootstocks</i>						
wild cherry						−0.40
<i>Prunus mahaleb</i>						−0.34
Cigánymeggy						−0.39
Altogether (n = 2100)	−0.44	−0.59	−0.48	−0.44	−0.28	−0.13

character of the trend of regression, Table 5 gives information. Accordingly, the negative character is of decisive importance; that is, the increase of pistil size is accompanied by the numerical reduction of stamina. We calculated 70 correlations per combination and year, and in 41 cases the value of  $r$  proved significant; the negative linear correlation was in full effect on the average of five years, varieties and yearly combinations.

The trends of pistil length and stamen number are regular and interconnected, which has a decisive influence partly on the fertility of reproductive

organs, partly — following from the above — on the productivity of the combinations.

On the basis of the facts Fig. 1 has probative force indicating the correctness of the above supposition; namely, that changes in the relative number of stamens have different effects on self-fertile and self-sterile sour cherry varieties. In the case of self-fertile varieties, the lower stamen number combinations produce the largest yields, while with the self-sterile ones it is just the other way round; the higher relative stamen number does not harm the chances of large yields (Fig. 1).

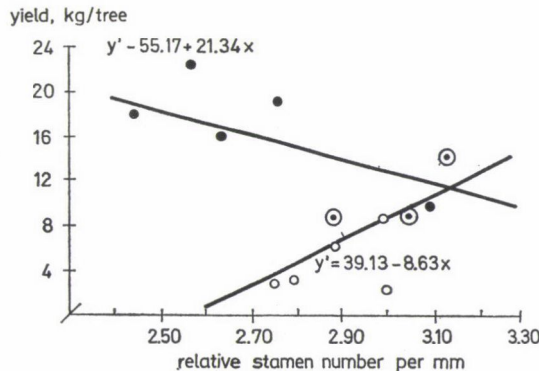


Fig. 1. Relationship between relative stamen number (average of 1976–1980) and fruit yield per tree (average of 1978–1980) in various stock-scion combinations of sour cherry (○). Self-sterile (Érdi nagygyümölcsű, Pándy C. 101),  $r = +0.770$ ; (⊙) partially self-fertile (Cigány-meggy C. 404); (●) self-fertile (Meteor korai, Újfehértói fürtös),  $r = -0.413$

The results of present study call attention to the importance of the morphological characters of sour cherry flowers, to the new aspect of stock effects. According to the experiences gained in practice, the “good” and “bad” rootstocks generally correspond to the higher or lower fertility of flowers. Morphological and statistical surveys of pistils and stamens have further enriched the work. The results agree with the conclusions of the literary works evaluated in the “Introduction”.

By the end of the present phase of research, important morphological data of flowers in many varieties have been collected, which together with the evidence of rootstock effect now supplied will be made complete — in accordance with the purpose — by the chemical control of fruit bud formation and flower organization. Namely, in the case of other stone fruit species, promising initial results have been attained in influencing the flower organization of self-sterile varieties, decreasing and increasing the chances of self-pollination, respectively, that is in the chemical control of fertility. The explanation of the physio-morphological mechanism of interference can be read in a recently published book (SURÁNYI 1980).



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### References

- BARG, T. (1958): Cytologische Untersuchungen an Sauerkirschen. *Gartenbauwiss.*, **23**, 200–208.
- FIELD, C. P. (1942): Low temperature injury to fruit blossom II. A comparison of the relative susceptibility and effect of environmental factors on three commercial apple varieties. Rept. East Mall. Res. Sta. for 1941, 29–35.
- LEMOINE, J. (1970): Anomalies des styles des anthères et des grains de pollen induites par diverses souches de virus des arbres fruitiers. Bordeaux, INRA Publ., 305–310.
- MALIGA, P. (1954): Untersuchungen über die Befruchtungsverhältnisse bei Weichselsorten. *Acta Agron. Hung.*, **4**, 105–149.
- MOLNÁR, L. (1963): A Pándy meggy virágzás- és ritkulásmenete, valamint gyümölcsfejlődése (Flowering and thinning course as well as fruit development of "Pándy" sour cherry). Duna–Tisza közti Mg-i Kísér. Int. Évk., 157–167.
- MORRISON, J., T. (1964): The stamen number of some fruit species and varieties grown at Morden, Manitoba. *Proc. Amer. Soc. Hort. Sci.*, **84**, 123–130.
- MURAWSKI, H.—ENDLICH, J. (1962): Beiträge zur Züchtungsforschung an Kirschen. II. Befruchtungsbiologische und embryologische Untersuchungen an der Sauerkirschen-sorte Köröser Weichsel. *Archiv f. Gartenbau*, **10**, 616–646.
- NYÉKI, J. (1974): Meggyfajták virágzása és termékenyülése (Flowering and fructification in sour cherry varieties). Candidate's dissertation, manuscript. Budapest.
- NYÉKI, J. (1975): Termékenyülési módszereink értékelése (Evaluation of methods of fertilization tests). *Kert. Egyet. Közl.*, 49–56.
- NYÉKI, J.—BRÓZIK, S. (1980): A meggy (The sour cherry). In: Nyéki, J. (ed.): The biology of flowering and fertility of fruit cultivars. *Mezőgazdasági Kiadó*, Budapest, 205–228.
- NYÉKI, J.—TÓTH, F. (1975): Meggyfajták és klónok virágainak morfológiai és fiziológiai sterilitása (Morphological and physiological sterility of flowers of cultivars and clones of sour cherries). *Bot. Közl.*, **62**, 287–298.
- NYÉKI, J.—TÓTH, F. (1976): Meggyfajták és klónok virágainak termőmorfológiai vizsgálata I. Pándy meggy klónok (Morphological studies by blossoms of sour cherry varieties and clones I. Clones of the variety Pándy). *Bot. Közl.*, **63**, 165–176.
- NYUJTÓ, F. (1971): Cseresznye és meggy alanyhatás kísérletek részeredményei (Partial results of experiments on stock effects as to cherry and sour cherry). *Szőlő- és Gyüm. term.*, **6**, 89–107.
- OLDÉN, E. J. (1965): Interspecific plum crosses. *Balsgard Fruit Breed. Inst.*, **1**, 1–58.
- PEJKIC, B. (1970): Frekventnost zaostavanja i konjugacija polivalentnih hromozoma u visnjoj sorte Kereske. *Arh. Poljop. Nauke*, **23**, 34–40.
- PHILP, G. L. (1933): Abnormality in sweet cherry blossoms and fruit. *Bot. Gaz.*, **94**, 820–851.
- PODDUBNAJA-ARNOLDI, V. (1964): Obscsaja embriologija pokrütoszemennüh rasztenij. A.N.Sz.Sz.Sz.R. Glavnüj Bot. Szad. Moscow, Izd. Nauka.
- RUDLOFF, C. F.—SCHANDERL, H. (1950): Die Befruchtungsbiologie der Obstgewächse und ihre Anwendung in der Praxis. Ulmer Verlag, Stuttgart.
- SCHANDERL, H. (1932): Untersuchungen über die Befruchtungsverhältnisse bei Stein und Kernobst in Westdeutschland. *Gartenbauwiss.*, **6**, 196–293.
- SCHANDERL, H. (1934): Über eine selbst sterile Spielart der Schattenmorelle. *Gartenbauwiss.*, **8**, 135–145.
- SURÁNYI, D. (1969–1972): Unpublished data, in SURÁNYI, D. (1979): Morfogenetikai tulajdonságok és összefüggések a *Prunoideae* alcsalád néhány nemzetiségének porzó- és termőtájában. Doktori értekezés, Budapest. (Morpho-genetical characters and relations in androecium and gynoecium of some *Prunoidea* genus. Doctoral dissertation, Budapest).
- SURÁNYI, D. (1976): Differentiation of self-fertility and self-sterility in *Prunus* by stamen number/pistil length ratio. *Hort. Sci.*, **11**, 406–407.
- SURÁNYI, D. (1977): Cseresznye- és meggyfajták virágsszerveződése (Flower organization of sweet- and sour cherry varieties). *Bot. Közl.*, **64**, 259–265.
- SURÁNYI, D. (1980): Masculin és feminin szexualitás egyensúlya a csonthéjasok virágában (Balance on stone fruit flowers by masculine and effeminite sexuality). In: Nyéki, J. (ed.): The biology of flowering and fertility of fruit cultivars. *Mezőgazdasági Kiadó*, Budapest, 34–42.

## CHANGES IN THE FREE PROLINE CONTENT OF GRAPE BERRIES DURING RIPENING

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Investigations carried out from 1976 to 1978 on quantitative changes taking place in the free proline content of fruits of four *Vitis vinifera* varieties (Rizling-szilváni, Ezerjő, Hárslevelű, Olaszrizling) under field conditions have led to the conclusion that proline shows an intensive accumulation in the course of ripening until, in the last sample — at the time of harvesting — reaching a level 25-50 times as high as in the initial phase of ripening. The accumulation of proline in the grape berry can be explained by the fact that it is derived from other amino acids through an intermediary metabolism; further, that the intensive formation of carbohydrates during ripening hinders its metabolization. Overdoses of nitrogen have ambiguous effects on the proline content. Out of the climatic factors it is the amount of annual precipitation and the intensity of light, rather than the alteration of temperature, that influence the change of the proline content.

### Introduction

Among works discussing the free amino acid composition of grape and must, a prominent place is occupied by studies on proline.

CASTOR (1953), LAFON-LAFOURCADE and PEYNAUD (1959) identified twelve amino acids in the must, of which proline and arginine were found to be present in the largest proportions. LAFON-LAFOURCADE and GUIMBERTEAU (1962) followed the changes in the free amino acids in the course of fruit ripening in the varieties Merlot and Cabernet Sauvignon, and pointed out that the leading role in the accumulation of amino acids was played by the proline. Similar observations were made by NASSAR and KLIEWER (1966), KLIEWER (1968, 1969), OUGH (1968), OUGH and ALLEY (1970), HACSIDZE and MATIKASVILI (1973).

DORER and MALNARIC (1978) consider the ratio of free amino acids/proline in the grape berry — that they call “proline index” — to be characteristic of the variety.

A number of results prove that the proline of all free amino acids is particularly responsive to changes in the environmental factors (MARH and SCSEBBAKOVA 1958, LAFON-LAFOURCADE and GUIMBERTEAU 1962). FLANZY and POUX (1965) revealed a higher concentration of proline in the must of grapes produced in a year with high temperatures during the growth season than in that obtained in a cool crop year.

COOMBE and MONK (1979) studied the effect of irrigation on the proline content of must. According to their opinion, the dry weather increases the



proline concentration of the must, but irrigation after a dry period does not change the proline level.

Studying the effects of an over-application of nitrogen nutrients, the researchers found that excessive nitrogen treatments mostly increased the amount of arginine but had no influence on the proline content (KLIEWER and COOK 1971, 1974, EWART and KLIEWER 1977, PEREZ and KLIEWER 1978, JUHÁSZ *et al.* 1980).

The experiment described here was aimed at following the changes in the free proline content of fruit during ripening in several wine-grape varieties and studying the effects of overdoses of nitrogen and potassium on the concentration of proline.

### Material and method

The quantitative determination of free amino acids, and of proline within, was carried out with a Mikrotechna AAA 881 type automatic amino acid analyser.

The conditions of the experiment, the set-up of nutrient treatments and methods employed were detailed in a previous publication (JUHÁSZ *et al.* 1984).

### Results

In the first harvest samples, little proline can be pointed out (0.5 mg/100 ml); but, by the time ripening is completed, the proline content may increase 25–50-fold, compared with its initial level (Table 1). The proline content of the last harvest samples reached a concentration of 16–18 mg/100 ml in 1976, 19–26 mg/100 ml in 1977, and 22–46 mg/100 ml in 1978, in the varieties “Rizlingszilváni” and “Ezerjő”. When expressed in percentage value (Table 2) proline represented 7–8.6, 11.5–15.8 and 10.6–20.0 per cent of the total amino acid content in 1976, 1977 and 1978, respectively. It is remarkable, however, according to results of analyses in Table 1, that the concentration of proline from July to September was higher in 1977 than in 1978.

Taking the climatic conditions in those years into consideration (Table 3) we find that, under the conditions of abundant precipitation and solar radiation in 1976, lower proline concentrations were found in the samples. The summer of 1977 was extremely dry, compared with the corresponding season of 1978, and the lower proline concentrations similarly occurred in the rainier period.

The higher proline content in the last harvest samples of 1978 may have been due to the lack of precipitation and the intensive solar radiation during September and October (Tables 1, 3). The conclusion drawn from all this is that the proline content correlates with the amount of precipitation throughout the year and during the vegetation period, and with the annual and monthly



**Table 1**  
*Changes in the proline content of must from control and high rate N- and K-fertilized vinegrape varieties*

Time	Control				High rate N- and K-fertilized	
	Ezerjő	Hárslevelű	Olasz-rizling	Rizling-szilváni	Rizlingszilváni	
					N	K
1976						
27 July	+	+	+	+	+	+
24 August	4.49	+	+	+	12.20	10.25
21 September	14.50	8.73	+	16.38	18.00	17.13
1977						
14 July	+	0.83	+	+	+	+
26 July	0.42	+	1.26	2.60	1.08	1.63
10 August	6.45	1.24	1.38	4.32	5.06	5.22
24 August	12.40	2.11	1.53	9.88	16.70	6.53
6 September	34.56	3.30	4.56	27.44	28.68	16.38
19 September	24.70	3.51	4.14	25.48	26.01	19.16
1978						
24 July	0.34	0.43	0.51	1.17	0.42	0.78
8 August	1.20	1.31	0.98	1.27	0.68	2.13
21 August	2.63	1.64	2.30	1.62	4.30	2.30
4 September	14.38	2.06	1.00	14.79	8.87	8.21
19 September	17.09	5.75	2.96	15.40	16.43	16.43
2 October	37.88	7.44	9.81	42.57	27.74	43.71
17 October	46.65	9.31	4.90	46.65	22.28	20.05

**Table 2**  
*Total free amino acids in the last harvest samples (mg/100 ml)*

Variety	1976	1977	1978
	21 September	19 September	17 October
Rizlingszilváni control	207.22	164.00	207.23
Rizlingszilváni N treatment	214.54	216.73	178.64
Rizlingszilváni K treatment	179.00	153.45	157.73
Ezerjő	128.24	166.63	165.14
Hárslevelű	116.93	69.80	85.67
Olaszrizling	120.86	101.41	99.50

number of sunshine hours, rather than with temperature change. As regards variety, the highest proline contents were found in the musts of "Rizlingszilváni" (29.5 mg/100 ml) and "Ezerjő" (28.6 mg/100 ml) on the average of three years. In the musts of "Hárslevelű" and "OlaszRizling", the concentration of proline was lower (7.18 and 4.5 mg/100 ml, respectively).

**Table 3**  
*Meteorological data from Szigetcsép*  
 (1976, 1977, 1978)

Month	Monthly and annual temperature mean, °C				Monthly and annual amount of precipitation, mm				Monthly and annual number of sunshine hours			
	1976	1977	1978	3-year average	1976	1977	1978	3-year average	1976	1977	1978	3-year average
I.	0.8	-0.1	0.1	0.2	25.0	41.0	22.0	29.3	56.0	45.0	63.0	54.7
II.	-0.1	4.2	0.4	1.5	3.0	53.0	28.0	28.0	93.0	88.0	57.0	79.3
III.	2.8	9.0	7.2	6.3	28.0	58.0	27.0	37.7	156.0	165.0	154.0	158.3
IV.	12.4	9.7	10.4	10.8	65.0	30.0	48.0	47.7	169.0	108.0	138.0	138.3
V.	17.1	17.1	14.4	16.2	20.0	45.0	77.0	47.3	199.0	203.0	158.0	186.7
VI.	20.4	21.2	18.7	20.1	30.0	13.0	64.0	35.7	207.0	204.0	217.0	209.3
VII.	23.1	20.8	20.2	21.4	105.0	54.0	69.0	76.0	211.0	197.0	233.0	213.7
VIII.	19.2	20.1	20.1	19.8	42.0	21.0	25.0	29.3	193.0	194.0	248.0	211.7
IX.	15.3	14.9	15.7	15.3	92.0	25.0	24.0	47.0	122.0	176.0	176.0	158.0
X.	11.6	12.0	11.6	11.7	80.0	16.0	21.0	39.0	99.0	165.0	180.0	148.0
XI.	7.0	5.7	2.2	5.0	30.0	61.0	16.0	35.7	53.0	81.0	16.0	50.0
XII.	0.1	-1.2	0.8	-0.1	99.0	29.0	28.0	52.0	37.0	32.0	37.0	35.3
Total	—	—	—	—	619.0	446.0	449.0	509.6	1595.0	1658.0	1677.0	1643.3
Average	10.8	11.1	10.1	10.7	—	—	—	—	—	—	—	—

As for the effect of nitrogen application on the proline content, it can be established that the must of the overtreated "Rizlingszilváni" contained a higher concentration of proline, according to the last harvest samples of 1976 and 1977; while in the samples of 1978 the proline level was lower, compared with the control (Table 1).

### Discussion

The observation that the proline level in the grape berry shows an intensive increase, with the advance of ripening, agrees with the results of other authors (OUGH 1968, COOMBE and MONK 1979). OUGH (1968) found 0.5 g/l proline in the ripe berry of "White Rizling", while COOMBE and MONK (1979) found 0.33–0.68 g/l proline in the must of the grape-vine variety Rizling.

According to our investigations the extent of proline accumulation in the grape berry changes from variety to variety. The analysis data concerning the variety "Rizlingszilváni" more or less agree with the results of the above authors.

During fruit ripening, changes in the sugar/acid ratio and the proline concentration are in positive correlation. The regression line expressing the

correlation as a function of the course of ripening ( $y = 0.68x + 2.73$ ) and the value of the correlation coefficient ( $r = 0.74$ ) are shown in Fig. 1; accordingly, the correlation is significant at  $P < 1\%$  level.

With the advance of ripening and at its last stage, in particular, the dehydration processes begin to play a dominant role, in the course of which an accumulation of sugars, increase in the respiration quotient and deamination of amino acids take place. As a result of the latter process, ammonia and  $\alpha$ -keto-glutaric acid accumulate. Subsequently the  $\alpha$ -keto-glutaric acid is used for the synthesis of glutamic acid and proline. The accumulation of proline occurs during photosynthesis, at which time the intensive formation of sugars prevents its oxidation (STEWART 1972).

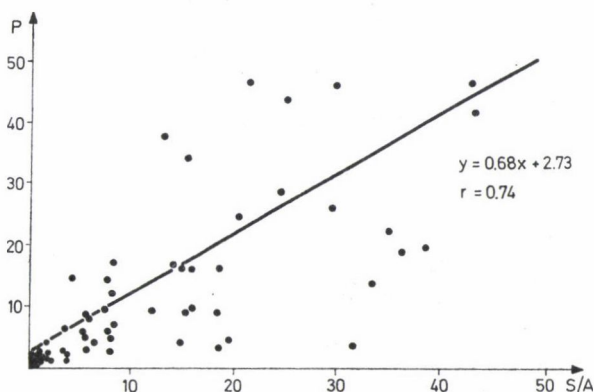


Fig. 1. Regression line ( $y$ ) expressing the correlation between the proline concentration of musts ( $P$ ) and the ratio of sugar to acid

All this accords with the conclusions drawn from earlier experiments, that the highest intensity of light (under which the activity of photosynthesis is also higher) and the concentration of proline correlate positively.

It has been pointed out that when the amount of carbohydrates in the leaf is reduced through translocation or respiration, the proline is metabolized in other amino acid and protein syntheses. The transformation of proline into glutamic acid produces NADPH; so, besides its role as a potential nitrogen pool, the proline represents a source of energy and reduction for the plant (DURZAN 1971). The accumulation of proline under stress (high intensity of light, lack of precipitation) can also be explained by this biological function, so important for the plant.

From the data of these three-year studies on the effect exercised by the nutrients on the quantitative change of proline, we can draw the conclusion that the influence of nitrogen and potassium on the proline content of the grape berry is doubtful.



## References

- CASTOR, J. G. B. (1953): The free amino acids of musts and wines. *Food Res.*, **18**, 139–145.
- COOMBE, B. G.—MONK, P. R. (1979): Proline and abscisic acid content of the juice of ripe Riesling grape berries: Effect of irrigation during harvest. *Amer. J. Enol. Vitic.*, **30**, 64–67.
- DORER, M.—MALNARIC, R. (1978): Proste Ammokisline v crnem grozdju in listju vinske trte (*Vitis vinifera* L.). *Farmaceutski Vestnik*, **29/2**, 93–110.
- DURZAN, D. I. (1973): Nitrogen metabolism of *Picea glauca*. V. Metabolism of uniformly labelled  $^{14}\text{C}$ -L-proline and  $^{14}\text{C}$ -L-glutamine by dormant buds in late fall. *Can. J. Bot.*, **51**, 359–369.
- EWART, A.—KLIEWER, W. M. (1977): Effects on controlled day and night temperatures and nitrogen on fruit-set, ovule fertility and fruit composition of several wine-grape cultivars. *Am. J. Enol. Vitic.*, **28**, 88–95.
- FLANZY, C.—POUX, C. (1965): Les levures alcooliques dans les vins. Protéolyse, protéogénèse (III). *Ann. Technol. Agr.*, **14**, 35–48.
- HACSIDZE, O. T.—MATIKASVILI, I. A. (1973): Izmenénije aminokislot v szoke vinograda pri szozrevanii. *Szad. Vinogr. i Vinodel. Moldavii*, **1**, 26–28.
- JUHÁSZ, O.—KOZMA, P.—POLYÁK, D. (1984): The effect of the supply with nitrogen nutrients on the free amino acid content of grapes. *Acta Agr. Hung. Acad. Sci.*, **33**, 3–17.
- KLIEWER, W. M. (1968): Changes in the concentration of free amino acids in grape berries during maturation. *Am. J. Enol. Vitic.*, **19**, 166–174.
- KLIEWER, W. M. (1969): Free amino acids and other nitrogenous substances of table grape varieties. *J. Food Sci.*, **34**, 274–278.
- KLIEWER, W. M.—COOK, J. A. (1971): Arginin and total free amino acids as indicators of the nitrogen status of grape vines. *J. Am. Soc. Hort. Sci.*, **96**, 581–587.
- KLIEWER, W. M.—COOK, J. A. (1974): Arginin levels in grape canes and fruits as indicators of nitrogen status of vineyards. *Am. J. Enol. Vitic.*, **25**, 111–118.
- LAFON-LAFOURCADE, S.—GUIMBERTEAU, G. (1962): Evolution des aminoacides au cours de la maturation des raisins. *Vitis*, **3**, 130–135.
- LAFON-LAFOURCADE, S.—PEYNAUD, E. (1959): Dosage microbiologique des acides amines des mouts de raisins et des vins. *Vitis*, **2**, 45–46.
- MARH, A. I.—SCSERBAKOVA, E. V. (1958): Aminokislotnűj szosztov vinogradnűh szokov. *Vinod. i Vinogr. S.S.S.R.*, **18**, 11–14.
- NASSAR, A. R.—KLIEWER, W. M. (1966): Free amino acids in various parts of *Vitis vinifera* at different stages of development. *Proc. Am. Soc. Hort. Sci.*, **89**, 281–294.
- OUGH, C. S. (1968): Proline content of vine grapes and wine. *Vitis*, **7**, 321–331.
- OUGH, C. S.—ALLEY, C. J. (1970): Effect of "Thomson Seedless" grape maturity on wine composition and quality. *Am. J. Enol. Vitic.*, **21**, 78–84.
- PEREZ, J. R.—KLIEWER, W. M. (1978): Nitrate reduction in leaves of grapevine and other fruit trees. *J. Am. Soc. Hort. Sci.*, **103**, 246–250.
- STEWART, C. R. (1972): Proline content and metabolism during rehydration of wilted excised leaves in the dark. *Plant. Physiol.*, **50**, 679–681.

## COMPLEX EVALUATION OF MAIZE HYBRIDS (*ZEAMAYS* L.) OF VARIOUS GENOTYPES

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Field trials, laboratory tests and feeding experiments with pigs were carried out with different maize genotypes (*Zeamays* L.). On the basis of these trials it can be established that the digestible energy yield correlated mainly to the crop yield while the digestible crude protein yield to the protein content and the digestibility of the protein. The digestible protein content depends rather on the crude protein content than on its digestibility. We found a positive correlation between the N-retention values and the protein content. The semi-dent (hard kernel) hybrids had generally a higher protein content and N-retention values than the dent, but there are exceptions, as well. The protein quality and quantity of the hybrids depended first of all on the specific combining ability of the inbred lines.

### Introduction

The evaluation of maize hybrids is based — with the exception of some experiments — on yield potential. The major share of maize is used as fodder; therefore, it is justified to take into consideration the reared amount of animal products, or the results of feeding trials.

It is generally known that the endosperm mutants, first of all the opaque ones, have a better nutritive value than the hybrids with normal endosperm, because of their more favourable composition (NELSON *et al.* 1965; VASAL 1971; ZUBER 1975; VASAL *et al.* 1980). As regards nutrient content, considerable differences were found among the hybrids with normal endosperm, too (SHOWALTER and CARR 1922; GENTER *et al.* 1957; DUDLEY and LAMBERT 1969; POLLMER *et al.* 1978).

Besides the crude nutrient content, NUSKERN *et al.* (1980) also examined the digestibility and the feed conversion rate. They wanted to know if there is any difference in the nutritive value between the early and the late hybrids. On the basis of feeding trials with pigs, they found the digestibility values of four hybrids with different growing season, as follows: dry matter 88.1–91.1%; crude protein 72.7–84.4%; crude fat 70.2–84.3%; crude fibre 41.1–60.5%; N-free extract 92.1–93.5%. The feed conversion rate varied between 3.29–3.44 kg/kg live weight gain. Meat quality was not influenced. The proportion of carcass meat varied between 40.3–40.6%.



There is a lack in our knowledge concerning any differences of practical importance in the nutritive value of hybrids with the same growing season but with different genotypes. Therefore, we made a complex evaluation of six hybrids in order to help plant breeding and the choice of hybrids for more economical animal production.

### Material and methods

We used hybrids bred by the Cereal Research Institute, Szeged. We selected hybrids that fit the needs of an average Hungarian farm (1500–2000 ha corn) in regard to agronomical values, growing season and genetic variability. The hybrids belong to two different growing season groups. (In Table 6 hybrids 1 and 2 are FAO 300, hybrids 3, 4, 5, 6 are FAO 500.) Both groups include semi-dent maize and dent maize hybrids, as well. The conclusions were based on the results of field trials, laboratory analyses, and digestibility and N-retention trials with pigs.

#### *Field experiments*

The yield potential of the hybrids was determined in a country-wide farm experimental network, including 100 farms in 1977, 1978 and 1979. The size of the plots was 1 or 2 ha. Apart from plant density (depending on the hybrid), and the date of harvest (28–30% moisture content), the applied production practices (seedbed preparation, fertilization, weed control) were similar to those applied on the other maize fields of the farms. More details can be found in the earlier publication of PINTÉR and SZIRBIK (1977). The corn used in the feeding trials with pigs was grown under the same circumstances (in the same field) in Lippó (county Baranya, South-Hungary) in 1979. The hybrids were harvested with an ear-maize picker at different dates, depending on the length of the hybrid's growing season.

The moisture content of the kernels at harvest was for the hybrids Sze TC 344, Sze SC 369, Sze MSC 515, Sze MSC 606, Sze MSC 565; 27, 29, 28, 30, 31%, respectively.

#### *Laboratory analyses*

The chemical composition (dry matter, crude protein, crude fat, crude fibre and ash) of the maize used in the feeding experiments was determined according to the standard MSz 6830. We determined the amino acid content by column chromatography with the analyser type Aminochrom (made by Labor MIM, Hungary).

#### *Digestibility and N-retention trials*

The harvested ear-maize was dried at room temperature to a moisture content of 15%. Afterward it was shelled. All samples taken to the chemical analysis and used in the feeding trials were prepared this way.

In the N-retention trials, we used weaned barrow piglets of Hungahyb hybrid pigs (bred by the Research Institute for Animal Breeding, Gödöllő).

The average weight of the piglets at the beginning of the experiments was  $20 \pm 2$  kg. A ten-day preliminary period was applied to accustom the piglets to the diet and to the metabolism cages.

After this preliminary, a first experimental period of seven days followed. During this time maize was practically the only diet (Table 1). This was followed by a five-day transition period, when the piglets were kept on a mixture of maize and extracted soybean meal. There followed a second experimental period of seven days, when maize and soybean meal mixture was fed to the piglets.

All hybrids were tested with 5 piglets each, in all the trials.

One half of the collected faeces was dried at 60 °C, the other was refrigerated till the chemical analysis. The excreted N was determined from the refrigerated sample, the other



**Table 1**

*The composition of the fodders used  
in digestibility and N-retention experiments*

Fodder component	Maize (first period)	Mixed fodder (second period)
	%	
Maize	96.0	72.0
Extracted soybean	—	24.0
AP-17	2.0	2.0
Kalcium	1.0	1.0
Salt	0.5	0.5
Premix XVII.	0.5	0.5
Total	100.0	100.0

excreted nutrients from the dried sample. The collected urine was covered with a thin film of toluol to prevent N losses during storage.

In the first experimental period, we determined the digestibility coefficients of the various maize hybrids, and in the second one the N-retention.

We determined the N-retention with a maize-soybean meal diet because, on one hand, corn is never fed alone in practice; therefore, the values determined exclusively for maize would have merely theoretical importance. On the other hand, the protein requirement of growing piglets cannot be satisfied by maize alone. Thus, the determined biological values would just express the efficiency of maize protein used for satisfying the protein requirement of subsistence.

The maize-soybean meal diet is very common in the nutrition of pigs, so the determined N-retention can be useful in practice. The experimental data also give us information about the way in which the amino acids of the two fodders supplement each other.

The hybrid Size SC 369 was used as standard (st). When determining the percentile deviation of the extreme values, the highest value was regarded as 100%.

## Results

The average yield values are shown in Table 2. There is a difference rising up to 13.6% among the hybrids. As is well-known, yield increases with growing season.

Calculating on the 87% dry matter basis, there is a significant difference in the crude fat and N-free extractives content, as compared with the standard (Table 3).

In connection with this, we have to remark that the crude protein content of our standard is between the extreme values. For this reason, the 8.0% deviation between the two extreme values, moreover all deviations above 5.5%, should be considered reliable. (This is the common standard error of sampling and examination.)

The digestibility values of nutrients differ significantly from the standard only in the case of crude fat (Table 4).

**Table 2**

*Air dried (15% H<sub>2</sub>O) maize yield on the basis of farm experiments (t/ha)*

Hybrids	1977	1978	1979	Average	Lippó 1979
Sze TC 344	—	7.20	7.04	7.12	7.08
Sze SC 369 (st)	7.01	7.32	7.01	7.11	7.51
Sze MSC 515	7.55***	8.33***	8.35***	8.08	8.69
Sze MSC 606	7.21*	8.65***	7.80***	7.89	8.99
Sze MSC 565	7.11	7.26	8.11***	7.49	9.65
KDC 22	—	—	7.55	7.55	7.90

\*, \*\*, \*\*\* = P<sub>5</sub>%, P<sub>1</sub>%, P<sub>0.1</sub>% as compared with Sze SC 369. In the case of "Lippó" and "Average" data, we did not use statistical analysis.

**Table 3**

*Chemical composition and nutritive value, calculated on 87% dry matter basis (g/kg)*

Hybrids	Organic matter	Crude protein	Crude fat	Crude fibre	Crude ash	Nitrogen-free extractives	Digestible energy (MJ)	Digestible crude protein
Sze TC 344	856.2	95.8	32.7	22.1	13.8	705.6	14.79	78.9*
Sze SC 369 (st)	857.9	93.3	30.7	22.2	12.1	711.7	14.61	74.1
Sze MSC 515	857.9	91.8	33.7	24.8	12.1	707.6	14.69	73.1
Sze MSC 606	854.9	88.1	41.7**	26.1	15.1	699.0**	14.76	70.0*
Sze MSC 565	856.5	93.9	30.8	26.6	13.5	705.2	14.41	72.7
K DC 22	857.9	94.9	41.6***	25.9	12.1	695.5***	14.81	75.5

\*, \*\*, \*\*\* = P<sub>5</sub>%, P<sub>1</sub>%, P<sub>0.1</sub>% as compared with Sze SC 369

**Table 4**

*Nutrient digestibility coefficients for pigs (%)*

Hybrids	Organic matter	Crude protein	Crude fat	Crude fibre	N-free extractives
Sze TC 344	92.37	82.34	80.18*	63.92	93.03
Sze SC 369 (st)	91.64	79.54	72.36	70.53	92.78
Sze MSC 515	91.72	79.64	78.16	73.66	92.39
Sze MSC 606	91.58	79.45	83.06*	71.13	92.06
Sze MSC 565	90.25	77.49	74.74	66.16	91.69
K DC 22	91.50	79.62	78.41	66.07	92.66

\* = P<sub>5</sub>% as compared with Sze SC 369

Table 5

*The N-retention values of the different hybrids (%) and the essential, or possibly essential, amino acids for pigs (g/kg) dry matter*

Hybrids	N-retention, %	Lysine	Histidine	Arginine	Threonine	Cystine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Phenylalanine
Sze TC 344	63.35	2.50	2.00	3.60	2.49	0.89	2.38	1.21	2.39	8.38	2.61	2.90
Sze SC 369 (st)	59.49	2.30	1.70	3.20	2.96	0.80	2.47	1.45	2.49	9.18	2.75	2.79
Sze MSC 515	48.19*	2.00	1.80	2.60	2.96	0.92	2.34	1.45	2.56	8.34	2.77	3.11
Sze MSC 606	49.57*	—	—	—	—	—	—	—	—	—	—	—
Sze MSC 565	55.84	2.30	1.80	3.00	2.50	1.17	2.10	1.65	2.44	8.32	2.89	3.22
K DC 22	59.89	2.70	2.00	3.50	3.05	1.13	2.72	1.47	2.67	8.53	3.39	3.22

\* = P<sub>5</sub>% as compared with Sze SC 369

It is worth mentioning that the gained crude fibre digestibility values are higher than those cited in the literature. This result may be due to the fact that the maize diet — consisting only of maize, mineral and vitamin supplements — contained less fibre than the requirement. This fact may have led to a good crude fibre digestibility. The data of N-retention experiments (maize-soybean meal mix diet) show differences in N-retention among the hybrids (Table 5).

### Discussion

The nutritive value of a fodder is determined, first of all, by its digestible energy and protein content. Both values show considerable differences, depending on the hybrid (Table 3). The nutrient yields also varied to a large extent (Table 6). The question can be raised, whether digestible energy and digestible protein yield were determined by yield or nutritive value.

In spite of the fact that we found significant differences in crude fat, N-free extractives and protein content, as well as in the digestibility of crude fat, the variation of the digestible energy yield was determined in 97.0% by the yield potential of the hybrid ( $r = 0.98^{***}$ ).

We did not find significant correlation between the digestible crude protein yield and the maize yield ( $r = 0.65$ ). This directs attention to the variability of protein content and quality.

We do not regard maize as protein fodder (as, for example, soybean) because of its relatively low protein content and because of the low biological



**Table 6**  
*The agronomical features and nutritive value of the hybrids*

Hybrid	Growing period*	Type of kernel	Yield, t/ha	Digestible energy yield, MJ/ha	Digestible crude protein yield, kg/ha
1. Sze TC 344 (GK21 × W117) — A 632	153	dent	7.12	105 305	561.8
2. Sze SC 369 (153R × SzV293)	156	semi-dent	7.11	103 877	526.9
3. Sze MSC 515 (GK17 × A632) — GK13	164	dent	8.08	118 695	590.6
4. Sze MSC 606 (GK5/1 × WF9 × A632)	165	dent	7.89	116 456	552.3
5. Sze MSC 565 (Oh43/301 × Oh43/H) — A632	167	dent	7.49	107 931	544.5
6. K DC 22 (WF9 × M14) — (K8 × K7)	169	semi-dent	7.55	111 816	570.0

\* Growing period = number of days from the shooting to the 28% moisture content stage

value of its protein. Despite this fact, maize plays a major role in the protein supply of the animals in Hungary. 70% of the grain fodder stock are maize. Therefore, 40% of the fodder protein originate from corn.

The correlation coefficient between the digestible crude protein and the crude protein content was  $r = 0.88^{**}$ . Between the protein digestibility coefficient and the digestible crude protein content, it was  $r = 0.72^*$ . These facts prove that the digestible crude protein content depended somewhat more on the protein content than on the digestibility.

One of the most important features of a protein is the amount retained in the animal organism.

There are important differences in amino acid content; that is, protein quality between hybrids with normal endosperm and those not bred for protein quality (Table 5). This mostly explains the considerable differences in the N-retention among the hybrids. (The difference between the extreme values was 23.9%.) The N-retention values may be influenced by the way in which the amino acid content of the hybrids and soybean-meal supplement each other. Though we do not know the amino acid composition of the hybrid Sze MSC 606, it can be established that there is a positive correlation between the lysine content of a hybrid and the N-retention.

The negative correlation between the protein quantity and quality is commonly known. Among nearly the same ecological circumstances as ours, GÁSPÁR (in KRALOVÁNSZKY 1975) tested 46 hybrids (the crude protein content varied 8–12%) and found the following correlation  $r = -0.55$ . Apparently, this contradicts the fact that we found a positive correlation ( $r = 0.84^*$ )

between the protein content and the N-retention. We calculated only with the results of the samples from the above-mentioned publication whose data fell into the protein content sphere of our hybrids in the trial (8.8–9.6%). Therefore, we found the correlation coefficient  $r = 0.44$  ( $n = 7$ ), instead of the earlier negative correlation. It refers to the fact that the negative correlation between protein quality and quantity exists only in a wide range of values. At the same time, we have to take into account that, in contrast to the cited literature data — where the biological values were determined only by chemical analysis — our N-retention values would be influenced not only by the amino acid composition of the maize, but also the supplementing effect of the two fodders. It might cause small differences, but cannot alter the tendency of correlation  $r = 0.92^{**}$  between the N-retention of fodder without extracted soybean and fodder with extracted soybean

Our data suggest the possibility of breeding hybrids with relatively high protein content and, at the same time, with high biological value. On the other hand, negative correlation is not necessary if a farm grows only a few hybrids.

The negative correlation between yield and protein quality is well known (Table 7). In accordance with the earlier research, we found a negative

**Table 7**  
*Correlation between the agronomical features  
and the nutritional value ( $r$ )*

	N-retention	Protein content	Yield
N-retention	1.00		
Protein content	0.84*	1.00	
Yield	-0.92**	-0.68	1.00
Growing period	-0.49	-0.30	0.66

\* = P5%; \*\* = P1%

correlation between the quantity and quality of protein, and the length of growing season. The low, statistically unreliable correlation coefficient means that there may be numerous exceptions. The possibility also exists to breed late hybrids with high protein content and good protein quality. In connection with the data, it is to be remarked that in our climate the low protein content and the worse protein quality of the late hybrids are, in most cases, due to improper maturity.

The moisture content of the samples collected for determining the nutritional parameters was approximately the same. Thus, the differences between hybrids have genetic origin.

The positive but insignificant correlation between yield and the length of the growing season calls attention to the fact that the influence of the growing season on the yield potential is not negligible; but the variability can often be higher within a single maturity group than between maturity groups.

Moreover, discussion is necessary to determine the correlation between properties can not be expressed with quantum, such as the type of the kernel, genetic composition, and the quality and quantity of protein. On the average, the N-retention values and protein content of semi-dent hybrids (Sze SC 369, and KDC 22) are higher than those of the dent ones (Tables 3 and 5). At the same time, it has to be mentioned that there are dent hybrids, such as Sze TC 344 and Sze MSC 565, whose protein parameters are better or not remarkably worse than that of semi-dent hybrids. Therefore, it cannot be generally accepted that the protein content is higher and the protein quality is better in the semi-dent (hard kernel) hybrids.

If we compared the protein content and the N-retention values of hybrids Sze MSC 515, Sze MSC 606 and Sze MSC 565 to the same values of hybrids Sze SC 369 and KDC 22, it could lead to the false conclusion that the inbred line A 632 inherits a lower protein content and N-retention value. At the same time, regarding both properties, the hybrid Sze TC 344 gave the best values among the hybrids containing the inbred line A 632 in the genetic basis. Consequently, the protein content and the protein quality of hybrids depend mainly on the specific genetic combination of inbred lines, and not on the presence of a certain line.

Further examinations are necessary to determine how N-retention is inherited.

### References

- DUDLEY, J. W. and LAMBERT, R. J. (1969): Genetic variability after 65 generations of selection in Illinois high oil, low oil, high protein and low protein strain of *Zea mays* L. *Crop Sci.*, **9**, 179.
- GENTER, C. F., EHEART, J. F. and LINKONS, W. N. (1957): Oil and protein relationships between corn inbred lines and their single cross progeny. *Agron. J.*, **49**, 283–285.
- KRALOVÁNSZKY, U. P. (1975): A fehérje probléma (Problem of protein). *Mezőgazdasági Kiadó*. Budapest.
- MSZ 6830–60 (1976). Takarmányok tápláléértékeinek megállapítása. Magyar Szabványügyi Hivatal. Budapest (Determination of fodders' feeding value. Hungarian Bureau of Standards. Budapest).
- NELSON, O. E., MERTZ, E. T. and BATES, L. S. (1965): Second mutant gene affecting amino acid pattern of maize endosperm proteins. *Science*, **150**, 1469–1470.
- NUSKERN, M., NOVOSLOVIC, A. and STEINER, Z. (1980): Hranjivo vrijednost nekik hibrida kukuruza u tovn svinja (The nutritive value of some corn hybrids in fattening of swine). *Zbornik Radova, Osijek*, **10**, 179–191.
- PINTÉR, L. and SZIRBIK, M. (1977): A kukorica (*Zea mays* L.) hibridek alkalmazkodóképességének vizsgálata [Examination of adaptability in maize (*Zea mays* L.) hybrids]. *Növénytermelés*, **26**, 433–442.
- POLLMER, W. G., EBERHARD, D., KLEIN, D. and DHILLON, B. S. (1978): Studies on maize hybrids involving inbred lines with varying protein content. *Z. Pflanzenzüchtung*, **80**, 142–148.



- SHOWALTER, M. F. and CARR, R. H. (1922): Characteristic proteins in high and low protein corn. J. Amer. Chem. Soc., **44**, 2019-2023.
- VASAL, S. K. (1971): CIMMYT's quality protein programme. Proc. of the 5th Maize Workshop. CIMMYT. Mexico. pp. 83-85.
- VASAL, S. K., VILLEGAS, E., BJARNASON, M., GELAW, B. and GOERTZ, P. (1980): Genetic modifiers and breeding strategies in developing hard endosperm opaque-2 materials. (Edited: Pollmer, W. G. and Phipps, R. H. Improvement of quality traits of maize for grain and silage use. Martinus Nijhoff Publishers. pp. 37-73.)
- ZUBER, M. S. (1976): Protein quality improvement in maize. Proc. 30th Annual Corn and Sorghum Research Conference. American Seed Trade Association. Washington D. C. pp.166-184.



## THE EFFECTS OF CHILLING TEMPERATURES ON THE CHLOROPLAST ULTRASTRUCTURE OF *CYNODON DACTYLON* AS AFFECTED BY GIBBERELIC ACID

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Low temperature discoloration of warm season perennial grasses is the result of a series of physiological disruptions referred to as chilling injury. Chilling injury occurs when air temperature is above freezing, but below 15 °C. Photosynthesis is one of the first physiological disruptions occurring in chill sensitive plants exposed to chilling temperatures. This disruption is closely followed by a deterioration or necrosis of the leaf tissue. It has been suggested that the photosynthetic reduction may be the result of excessive starch grain accumulation in the chloroplasts. It has also been suggested that exogenous gibberellic acid (GA<sub>3</sub>) may dissipate accumulated starch by stimulating amylase activity. This theory was tested by exposing GA<sub>3</sub> treated and non-treated Pee Dee (chill sensitive) and Ormond (chill tolerant) bermudagrass [*Cynodon dactylon* (L.) Pers.] to chilling temperatures and then examining their chloroplast ultrastructure with a transmission electron microscope.

Starch grain accumulation in the bundle sheath chloroplasts of both non-treated bermudagrass cultivars at the end of the initial chilling day were similar and always substantially lower than the pre-stress levels. Ormond treated with GA<sub>3</sub> showed a reduction in the number of starch granules in the bundle sheath chloroplasts when compared to non-treated Ormond turf. However, there was no reduction in the number of starch grains in the bundle sheath chloroplasts from GA<sub>3</sub> treated Pee Dee when compared to non-treated Pee Dee plants. Mesophyll and bundle sheath chloroplast ultrastructure, including granal stacking, lamellae system and peripheral reticulum when present, appeared normal in both bermudagrass cultivars after 18 hours of chilling stress.

### Introduction

Low temperature discoloration and subsequent winter dormancy of warm season perennial grasses occur when soil and air temperatures are above freezing but below 15 °C. The discoloration is the result of physiological disruptions referred to as chilling injury.

A significant reduction of photosynthesis occurs when chill sensitive plants are exposed to chilling temperatures (5 to 15 °C) (MILLER 1960, LUDLOW and WILSON 1970). The disruption of photosynthesis may be the result of ultrastructure changes in the chloroplasts. For example, TAYLOR and CRAIG (1971) reported extensive swelling and subsequent thylakoid disruption in the chloroplasts of *Sorghum* (chill sensitive) when exposed to 10 °C nights and 25 °C days. In the same study, chloroplasts of *Paspalum* (chill tolerant) also exhibited significant swelling, but the internal membrane system was held much more intact. It has been proposed that excessive starch accumulation in



chloroplasts due to chilling temperatures is responsible for the gross swelling of chloroplasts and ultimately the loss of photosynthetic capability. HILLIARD and WEST (1970) examined young leaf tissue from *Digitaria decumbens* Stent. which had been exposed to continuous 12-hour 30 °C nights and leaf tissue from plants which received continuous 12-hour night temperatures of 30 °C followed by one 12-hour night at 10 °C. Mesophyll chloroplasts contained large amounts of starch grains at the end of the light period. Starch grains were absent from the chloroplasts at the end of the dark period at 30 °C, but chloroplasts from plants receiving the low night temperatures of 10 °C contained starch grains similar to those found at the end of the light period. Repeated 10 °C exposure during dark periods resulted in continued starch accumulation. After only one night exposure to 10 °C, this starch accumulation may have accounted for the swollen appearance of the chloroplasts and may have interfered with photosynthesis. The implication is that the severe reduction in growth and photosynthesis of *Digitaria decumbens* following chilling night temperatures may result from failure to translocate photosynthetic products from the mesophyll chloroplasts. KARBASSI *et al.* (1972) suggested that the failure of starch to be translocated from the chloroplasts of *Digitaria decumbens* may be due to a low temperature sensitivity of some starch degrading enzymes. They found the amylolytic activities of leaf extracts from *Digitaria decumbens* to be very low following 10 °C night temperatures. To further test this, CARTER *et al.* (1973) treated *Digitaria decumbens* with gibberellic acid (GA) and subjected the plants to 30 or 10 °C nights. GA treatments of plants subjected to 10 °C increased the starch degrading enzyme activity to the level of activity found in 30 °C untreated (control) plants. GA treatment of plants at 10 °C decreased sucrose and starch levels in the leaves compared to chilled leaves without GA. KARNOK and BEARD (1983) found a differential photosynthetic response of chill sensitive and chill tolerant cultivars of *Cynodon dactylon* and *Stenotaphrum secundatum* treated with GA. *Cynodon* cultivars showed a significant increase in photosynthesis 10 hours after GA application. The *Stenotaphrum* cultivars either showed no photosynthetic response or a significant reduction.

### Materials and methods

Mature sods, 10 cm in diameter by 7.5 cm deep, of two bermudagrass [*Cynodon dactylon* (L.) Pers.] cultivars, Pee Dee (chill sensitive) and Ormond (chill tolerant) were obtained from the Texas A & M Turfgrass Field Laboratory. After washing soil from the roots, the sod was placed in an 8.75 cm diameter by 10 cm deep plastic pot containing 100% mortar sand. The plant material was maintained outdoors approximately 4 weeks prior to chilling treatment. The average day/night temperatures during this period were 35/25 °C. The sod was irrigated with distilled water as needed to prevent wilt and received a complete Hoagland's (1950) nutrient solution applied as a drench twice weekly. The plant material was clipped twice weekly at 1.0 cm with the clippings being removed.

A two-week acclimation period involved the transfer of two randomly selected pots of each cultivar from outdoors to a modified Sherer environmental growth chamber. The modifica-

tion involved replacement of the standard fluorescent-incandescent lighting with an autonomous light bank consisting of three Sylvania 1000 W Super Halide lamps. This light source provided  $1400 \mu \text{E m}^{-2} \text{Sec}^{-1}$  PAR radiation at the turf surface. A 12-hour photoperiod and a day/night temperature of 32/27 °C were used throughout the acclimation period.

Following the acclimation period, and at the beginning of the dark period, the temperature was lowered 3 °C per hour until leaf temperature reached 5 °C. GA<sub>3</sub> at 62.5 g/ha was applied to one pot of each cultivar two hours after commencement of the next light period at a 10 °C leaf temperature. A day/night air temperature of 7/5 °C and a 12 hour photoperiod were maintained throughout the remainder of the study.

Samples were taken from the middle region of the second youngest fully expanded leaf. Leaf samples from six tillers of each cultivar (both GA<sub>3</sub> treated and non-treated) were taken under the following conditions: (1) at the end of the last pre-stress photoperiod, (2) immediately following the dark period at the pre-stress temperature, (3) immediately following the dark period after the initial 5 °C night, and (4) ten hours after commencement of the photoperiod (eight hours after GA<sub>3</sub> application) at chilling temperatures.

Leaf sections, 1 × 4 mm, were (a) cut under 5 °C and placed in 2.5% glutaraldehyde and 0.5% acrolein in 0.1 M S-collidine buffer (pH 7.2) containing 0.01 M CaCl<sub>2</sub>; (b) vacuum-infiltrated for 15 minutes; and (c) postfixed for one hour in one part of the above solution mixed with one part OsO<sub>4</sub> (2%) in 0.1 M S-collidine. The specimens were then dehydrated for 15 minutes in each concentration in a graded ethanol series (50, 70, and 95%). This was followed by a 15 minute further dehydration in 100% ethanol, three separate times. The material was then transferred to 100% propylene oxide for 30 minutes. The samples were then infiltrated with a propylene oxide resin series 2 : 1 and 1 : 1 for 15 minutes each, followed by 30 minutes in a 1 : 2 propylene oxide resin mixture. The specimens were then preembedded in pure resin for 60 minutes followed by permanent embedment in Araldite. Sections were post-stained for five minutes in alcoholic uranyl acetate and lead citrate, and examined with a transmission Hitachi 11-U electron microscope. Chilling treatment, as well as tissue sampling and preparation were replicated two times.

## Results and discussion

The mesophyll and bundle sheath chloroplasts of both cultivars contained no starch grains after 12 hours in the dark at 27 °C. However, at the end of the initial dark period at chilling temperatures, starch grains were abundant in the bundle sheath chloroplasts of both bermudagrass cultivars (Fig. 1). There was no starch grain accumulation in the mesophyll chloroplasts of either

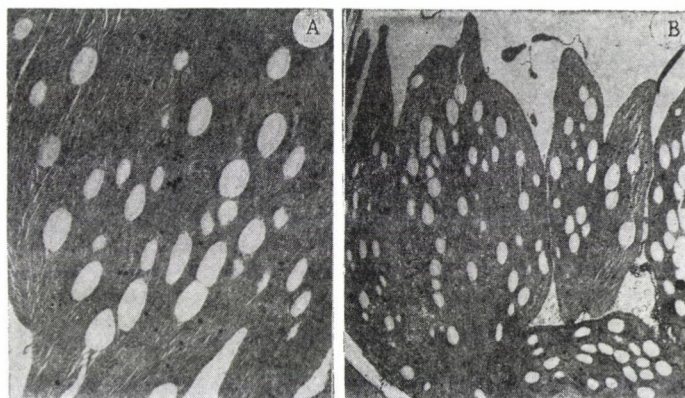


Fig. 1. Transmission electron micrographs of Pee Dee (A) ( $\times 12\,400$ ) and Ormond (B) ( $\times 8400$ ) at the end of the initial chilling



cultivar and no apparent granal reduction or membrane disruption in either type of chloroplast. The ability of bermudagrass to accumulate starch in the bundle sheath chloroplasts rather than the mesophyll chloroplasts has been documented by RHOADES and CARVALHO (1944). HILLIARD and WEST (1970) reported that pangolagrass (*Digitaria decumbens* Stent.), which stores starch in the mesophyll as well as in the bundle sheath chloroplasts contained prominent starch grains in mesophyll chloroplasts after one nocturnal exposure to 10 °C.

Turfs of both cultivars which did not receive the GA<sub>3</sub> treatment contained starch in the bundle sheath chloroplasts at the end of the 10 °C light period following the initial dark period of chilling temperature (Fig. 2). However, the number of starch granules accumulated was always less than half the number observed in the bundle sheath chloroplasts at the end of the last pre-stress photoperiod (Fig. 3). Granal stacking as well as the rest of the stroma lamellae appeared to be intact and normal in both mesophyll and bundle sheath chloroplasts. TAYLOR and CRAIG (1971) reported stromal swelling, a decrease in the thylakoid intraspaces widths, and a reduction in granule stacking in the mesophyll chloroplasts of *Sorghum* after 1.5 days at 10 °C. The lack of agreement between these two studies is probably due to length of exposure to chilling temperatures. The *Sorghum* was exposed to 10 °C for 36 hours, whereas the two bermudagrass cultivars used in this study had received approximately 18 hours of chilling temperatures.

Pee Dee bermudagrass treated with GA<sub>3</sub> two hours after commencement of the first photoperiod following the initial dark period of chilling temperatures, did not show a reduction in starch grain accumulation in the bundle sheath chloroplasts ten hours after the GA<sub>3</sub> was applied (Fig. 4). However, Ormond bermudagrass had a definite reduction in starch granule accumulation in the bundle sheath chloroplasts ten hours after GA<sub>3</sub> was applied (Fig. 4). Starch grains were completely absent in many of the bundle sheath chloro-

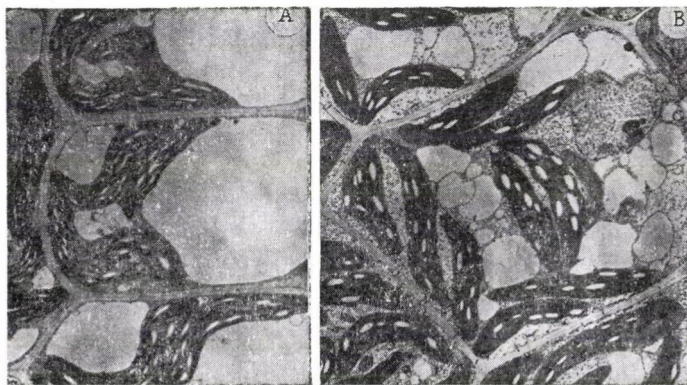


Fig. 2. Transmission electron micrographs of Pee Dee (A) and Ormond (B) without GA<sub>3</sub> treatment at the end of the initial 10 °C photoperiod ( $\times 6000$ )





Fig. 3. Transmission electron micrograph of a typical bundle sheath chloroplast of Pee Dee and Ormond bermudagrasses at the end of the last pre-stress photoperiod ( $\times 9200$ )

plasts, whereas the Ormond bermudagrass not receiving the  $GA_3$  treatment had several starch granules in every bundle sheath chloroplast. The apparent reduction of starch granules in the bundle sheath chloroplasts of Ormond treated with  $GA_3$  could be explained by an increase in amylase activity (CARTER *et al.* 1973). However, this starch grain reduction did not occur in the chloroplasts of Pee Dee. This lack of response to  $GA_3$  may be due to the rate used. There was no starch accumulation in the mesophyll chloroplasts of either bermudagrass cultivar. The ultra-structure of both mesophyll and bundle sheath chloroplasts appeared normal in both cultivars.

The highest level of starch granule accumulation in the chloroplasts of either cultivar occurred at the end of the last pre-stress photoperiod. All the bundle sheath chloroplasts were tightly packed with starch granules. While exposed to chilling temperatures, the starch granule accumulation in the bundle sheath chloroplasts from either cultivar (treated or not-treated) was always substantially less than the pre-stress levels. There were no obvious differences between the non-treated cultivars at the end of the initial chilling night, or initial chilling day.

The results from this study suggest that a rapid reduction in net photosynthesis following a chilling night may not be the result of excessive starch accumulation that causes mechanical disruption in the bundle sheath chloroplasts. TAYLOR and ROWLEY (1971) reported a dramatic reduction in photo-



Fig. 4. Transmission electron micrograph of Pee Dee (A) ( $\times 16\,900$ ) and Ormond (B) ( $\times 16\,600$ ) bundle sheath chloroplasts treated with  $GA_3$  at the end of the initial  $10^\circ\text{C}$  photoperiod

synthesis of *Sorghum* (Hybrid NK, 145) immediately following exposure to  $10^\circ\text{C}$ . However, chloroplast ultrastructure changes did not occur until the plant material was exposed to  $10^\circ\text{C}$  for at least 1.5 days. Therefore, the results of these studies indicate a more significant primary factor is responsible for the initial reduction in net photosynthesis of plants exposed to chilling temperature.

### References

- CARTER, J. L., GARRARD, L. A. and WEST, S. H. (1973): Effect of gibberellic acid on starch degrading enzymes in leaves of *Digitaria decumbens*. *Phytochem.*, **12**, 251–254.  
 HILLIARD, J. H. and WEST, S. H. (1970): Starch accumulation associated with growth reduction at low temperatures in a tropical plant. *Science*, **168**, 494–496.  
 HOAGLAND, D. R. and ARNON, D. I. (1950): The water culture method for growing plants without soil. *California Agr. Exp. Sta. Cir.*, 347.  
 KARBASSI, P., WEST, S. H. and GARRARD, L. A. (1972): Amyolytic activity in leaves of a tropical and a temperate grass. *Crop. Sci.*, **12**, 56–60.

- KARNOK, K. J. and BEARD, J. B. (1983): Effects of gibberellic acid on the CO<sub>2</sub> exchange rates of bermudagrass and St. Augustinegrass when exposed to chilling temperatures. *Crop Science*, **23**, 514-517.
- LUDLOW, M. M. and WILSON, G. L. (1970): Photosynthesis of tropical pasture plants. I. Illuminance, carbon dioxide concentration, leaf temperature, and leaf-air vapor pressure difference. *Aust. J. Biol. Sci.*, **24**, 449-470.
- MILLER, V. J. (1960): Temperature effect on the rate of apparent photosynthesis of seaside bent and bermudagrass. *Proc. Amer. Soc. Hort Sci.*, **75**, 700-703.
- RHOADES, and CARVALHO, A. (1944): The function and structure of the parenchyma sheath plastids of the maize leaf. *Bull. Torrey Bot. Club*, **71**, 335-346.
- TAYLOR, A. O. and CRAIG, A. S. (1971): Plants under climatic stress. II. Low temperature, high light effects on chloroplast ultrastructure. *Plant Physiol.*, **47**, 719-725.
- TAYLOR, A. O. and ROWLEY, J. A. (1971): Plants under climatic stress. I. Low temperature, high light effects of photosynthesis. *Plant Physiol.*, **47**, 713-718.





## PURIFICATION AND SOME PROPERTIES OF MYOSIN PREPARED FROM ROOT TIPS OF MAIZE (*ZEA MAYS* L.) SEEDLINGS

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The root tips of all angiospermae consist of the division of cells of meristematic tissue. The idea has arisen that myosin must be present in all proliferative tissues, because the cytokinetic movements occur on myosin filaments through different transmission mechanisms, depending on their origin and the function of the myosin. We set out to check this idea. Myosin was isolated from the root tips of maize seedlings. This myosin was purified through gel-filtration and phosphorylated. It contains basic amino acid phosphates. Myosin in root tips functions mainly at the expense of P-Arg. This myosin is itself able to build up a filamentous system.

### Introduction

Concerning plant cells, it appears increasingly important to learn more about the functions of the macrotubules, microtubules and microfilaments. The dynamic approach to ultrastructures is currently spreading.

Recent investigations have demonstrated that the main proteins of muscle, actin and myosin, are found not only in muscle cells, but also in most types of eukaryotic cells, ranging from vertebrates to amoebae (CLARKE and SPUDICH 1977). WAGNER (1979) believes that the role of actomyosin is also general in the phenomena of movement in the plant kingdom too.

Mostly the lower plants, such as algae and slime fungi, have been investigated. As models, a myosin B-like protein and actomyosin have been found in *Nitella flexilis*, and actin, myosin and actomyosin in *Physarales* (KATO and TONOMURA 1977, BRITZ 1979).

Few data are available on the presence of actin and myosin in higher plants. Actin has been isolated from soybean seedlings (METCALF III *et al.* 1980), and a tubulin-like protein found in *Phaseolus aureus* (RUBIN and COUSIN 1976). Actomyosin has been isolated from root tips of *Phaseolus vulgaris* (JACKSON and DOYLE 1977). VAKEX *et al.* (1978) found actin and myosin in parenchyma tissue of fruits of *Lycopersicum esculentum*.

## Material and method

1–1.5 cm root tips (300 or 500 g) from seedlings of maize hybrid SZE DC 384 germinated in the dark were collected in a glass vessel containing ground ice, and stored at 0 °C until use. The root tips were minced in a precooled grinder, homogenized in a triple volume of extracting solution [0.5 mM KCl, 25 mM Tris-HCl (pH 7.6) and 2 mM mercaptoethanol] and centrifuged at  $5000 \times g$  for 30 min at 0 °C. The extraction was repeated and the collected supernatants were dialysed first against distilled water and then against 25 mM KCl, the pH being kept at 7.15 with a small amount Tris-HCl buffer. Buffers containing phosphate were avoided. The precipitate was collected by centrifugation ( $20\,000 \times g$ , 30 min at 0 °C), dissolved in a minimal volume of 2 M KCl–25 mM Tris-HCl (pH 7.6) and diluted to 0.5 mM KCl, followed by centrifugation ( $16\,000 \times g$ , 30 min at 0 °C).

The myosin was precipitated from the supernatant by dialysis and recovered by centrifugation. The pellet was dissolved as before and the solution was centrifuged. The subsequent purification was as described by FAZEKAS *et al.* (1979). The purification was completed by means of DEAE-cellulose treatment and centrifugation ( $105\,000 \times g$  for 90 min at 0 °C) to remove RNA, actin and nucleotide traces, with gel-filtration on a Sepharose 4B column. The tubes relating to the first peak were taken as containing myosin, and the LC fraction too (light chains + accompanying proteins + P-lipids) was sometimes collected.

The protein content of the product was determined by the micro-biuret method of GOA and SCAN (1963), using salt- and lipid-free myosin (measured gravimetrically after the removal of moisture at 105 °C) as internal standard, via ultra-violet absorbance at 280 nm (Fig. 1).

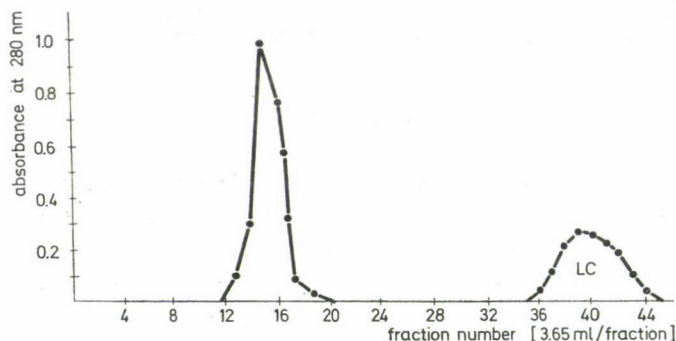


Fig. 1. Uv absorbance of gel-filtered myosin

Gel-filtration of root tip myosin from maize seedlings was performed on a Sepharose 4B column ( $1.85 \times 65$  cm). The column was equilibrated with 0.5 M KCl, 8 mM  $\text{NaHCO}_3$  and 0.5 mM DTT, and the sample was applied in a total volume of at most 4 ml. The elution was done with the same buffer and the protein content was followed via ultra-violet absorbance at 280 nm. The tubes of the first fraction were pooled as myosin, which was applied for the determination of ATPase activity, the formation of myosin filaments, light and electron microscopy and the preparation of basic amino acid phosphates.

The lipid content was removed by the method of FOLCH *et al.* (1957), through double extraction with  $\text{CH}_2\text{MeOH}$  (2 : 1 by vol) and the P-lipid content of the total product was determined, attention being paid to the presence of inorganic P-traces.

P-content determinations were performed on the inorganic residues of the samples, after decomposition with  $\text{cc HNO}_3$ , by the method of FISKE and SUBBAROW (1925), except that the final reduction was achieved by adding 1% ascorbic acid (0.25 ml for 2.5 ml final volume) according to LOWRY and LOPEZ (1942). The method is suitable for the determination of 0.02–1.5 micromol P in the samples.

The amount of acid-labile phosphate of N-P type was studied through 0.42 mM  $\text{Cu}^{2+}$  treatment (kept at 0 °C over night) in order for the Cu-protein chelate complex to form and

### Abbreviations:

$\text{CH}_2$  = chloroform, DTT = dithiothreitol, HC = heavy chain, LC = light chain,  $\text{MeOH}$  = methanol, P = macroerg organic phosphate, Pi = inorganic phosphate.



precipitate as a pellet. The complex was separated from the supernatant by centrifugation, both parts being used for P determination. Internal controls were samples without protein and another without copper. Acetone precipitation was applied to check complete precipitation of protein and others, for the correction of colour intensity.

The myosin obtained from the root tips could be phosphorylated with a suitable phosphorylating mixture, as described in Table 1.

Table 1  
*Phosphorylation of gel-filtered root tip myosin of maize seedlings*

Sample	mol P/mol myosin
Gel-filtered myosin (control)	15.4
Internal control (omitted ATP)	5.38
Phosphorylated samples (no treatment)	20.8
pretreated with $\beta$ -indoleacetic acid (1 mM)	17.4
pretreated with colchicine (1 mM)	12.4
pretreated with $\text{Cu}^{2+}$ (0.42 mM)	7.1

The phosphorylation ability of myosin was determined in a two-stage incubation system in 2 ml final volume containing: 0.22–0.5 mg myosin, 100 mM KCl, 25 mM Tris-HCl (pH 7.4), 0.1 mM DTT (dithiothreitol), 12 mM  $\text{MgCl}_2$ , 0.15 mM ATP, 60 mM NaCl and 0.5 mM  $\text{CaCl}_2$ . 3-Indoleacetic acid, colchicine and  $\text{CuCl}_2$  were sometimes added prior to incubation.  $\text{MgCl}_2$  and ATP were added as Mg-ATP, followed by a  $\text{CaCl}_2$ –NaCl mixture. The mixture was incubated at 25 °C for 3 min and terminated with 5 volumes of ice-cooled acetone; after 6 hours' standing at 0 °C, the protein was pooled by centrifugation.

The superfluous nucleotides and Pi were removed with washing solution (20 mM KCl, 10 mM NaCl, 2 mM  $\text{MgCl}_2$ , 0.1 mM  $\text{CaCl}_2$  and 5 mM  $\text{NaHCO}_3$ ; one volume of this solution in 2 volumes of ethanol), washed 6 times with the washing solution and once more with the solution (ethanol omitted), and the procedure was completed by the removal of lipids with a  $\text{CH}_2\text{MeOH}$  (2 : 1 by vol) mixture. The lipid-free protein and lipid fractions were used for P determination. The P-contents were calculated for a 478 000 D lipid-free preparation. The internal control without ATP was subjected to the washing procedure in parallel with the other samples, to check the efficiency of the washing.

Amino acid phosphates from the alkaline hydrolysates were separated and determined, as described in Fig. 2; and their specific reactions were similar to those described by FAZEKAS *et al.* (1981). The P-content was examined in an aliquot of every effluent tube, as described FAZEKAS *et al.* (1980).

## Results and discussion

Root tips from maize seedlings yield 16.5% dry matter containing 20.2 micromol P/g wet mass or 128 micromol P/g dry mass. When the pooled root tips were stored in the frozen state and then used for the preparation of myosin, as a consequence of the increased proteolysis and phosphatase activity, the diffusible P was dispersed in the surrounding solution.

The last phase of the purification was performed by gel-filtration, as shown in Fig. 1. The tubes relating to the first peak were collected as myosin from 100 g wet root tips. The ultra-violet spectrum was flat and uncharacteristic. 1 mg/cm<sup>3</sup> myosin gave an absorbance (A) 0.445 at 280 nm, with  $A_{280}/A_{260} = 0.905$ .

The gel-filtered product contained on average 28.7 mol P/mol, calculated for 478 000 Dalton (D) as probable molecular mass. A significant amount of P-lipids (12.6 mol) was found in the gel-filtered preparation by double extraction separation with a  $\text{Chl-MeOH}$  (2 : 1 by vol) mixture.

The remaining P (15.4 mol) was bound covalently with different bond strengths, linked to several basic amino acid residues.

Most of the P appeared to be acid-labile phosphate with N-P type linkage. On the action of 0.42 mM  $\text{Cu}^{2+}$ , about 4–6 M P was released, with additional P liberated by 1 mM  $\text{Cu}^{2+}$ , which formed chelate complexes with the basic amino acids. A little residual P released from its myosin on alkaline hydrolysis, showing it to be present in ester bonds.

The tubes relating to the light chain range contain 15 000–30 000 D proteins. When the P-content was determined in this fraction, a significant amount of P (4.75 M) was found in the lipid-free protein, compared with 15.4 M P for the control and 15 M P for the internal control. The P incorporation was moderated in myosin pretreated with 3-indoleacetic acid or colchicine, and prevented with  $\text{Cu}^{2+}$ .

Part of the myosin was applied for the isolation of basic amino acid phosphates. The bulk of these phosphates is P-Arg. This is seen in Fig. 2.

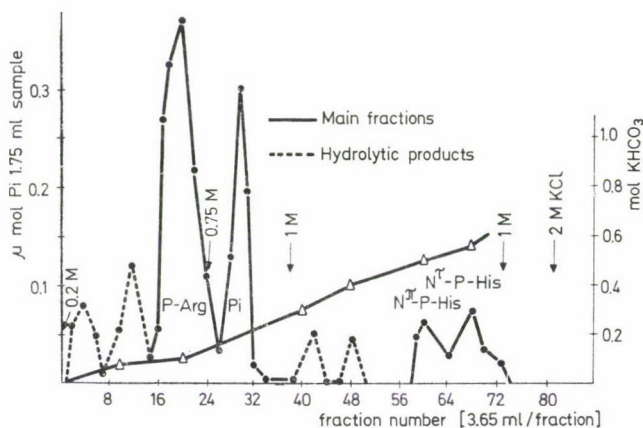


Fig. 2. Basic amino acid phosphates from hydrolysed myosin

Basic amino acid phosphates were separated from the alkaline hydrolysate of the root tip myosin of maize seedlings on a Dowex 1 X8 column ( $0.9 \times 6$  cm). 18 mg lipid-free protein was hydrolysed in 3 M KOH for 10 h at 105 °C in a sealed Pyrex glass ampule, applied to the column diluted to 0.01 M KOH, and chromatographed by a linear gradient step method. A mixing chamber with 160 ml capacity was filled with 0.01 M  $\text{KHCO}_3$ , and the reservoir with 120 ml 0.2 M  $\text{KHCO}_3$ . The concentration in the reservoir was increased at the arrows to 0.75 and 1 M  $\text{KHCO}_3$ , at the mark 1 M  $\text{KHCO}_3$  was applied directly without a mixing chamber or electromagnetic stirrer, and the concentration was finally changed to 2 M KCl. Under the applied conditions, the neutral and basic amino acids were not bound on the Dowex resin, but only the dicarboxylic amino acids, basic amino acid phosphates and inorganic P. The P content in the effluent was followed via a modified molybdate reaction in the presence of perchloric acid. Only the tubes containing P were collected. After lyophilization, these applied for paper and thin-layer chromatographic control and specific amino acid reactions.



The root tip myosin is itself able to form a filamentous systems (Figs 3, 4 and 5).

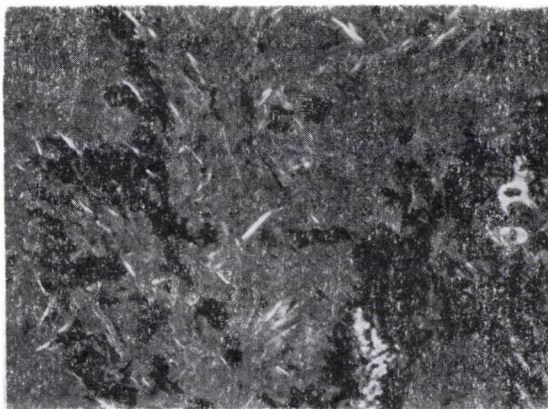


Fig. 3. Polarizing micrograph of filamentous structure of root tip myosin ( $\times 300$ )

The gel-filtered myosin formed a filamentous system on storage at  $0^{\circ}\text{C}$  in the presence of  $1\text{ mM K[Au(CN)}_4\text{]}$ . This was post-contrasted with  $\text{AuCl}_3$  and treated with  $\text{AgCl}$  to show anisotropy. The clear filamentous aggregates are birefringent and consist of subfilaments.

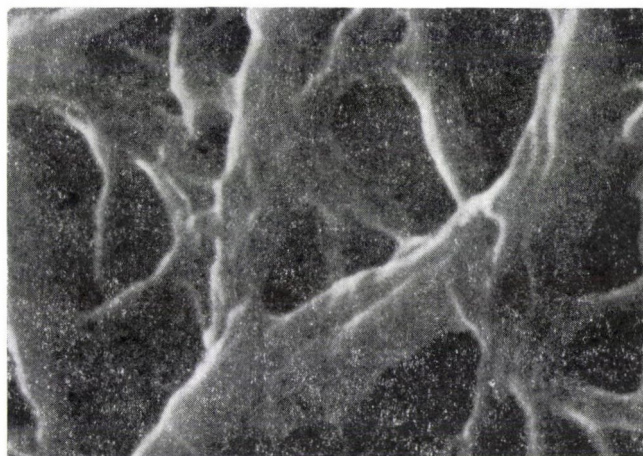


Fig. 4. Scanning electron micrograph of root tip myosin ( $\times 7300$ )

The filaments show the heads of myosin. The surface of the filaments display numerous heads, both single and double. On long storage, the filaments tend to form rough aggregations. When the surface of the filament bundles seems smooth, the heads are merged in their surroundings; in the process more and more inorganic P is released into the solutions of the treated preparation.



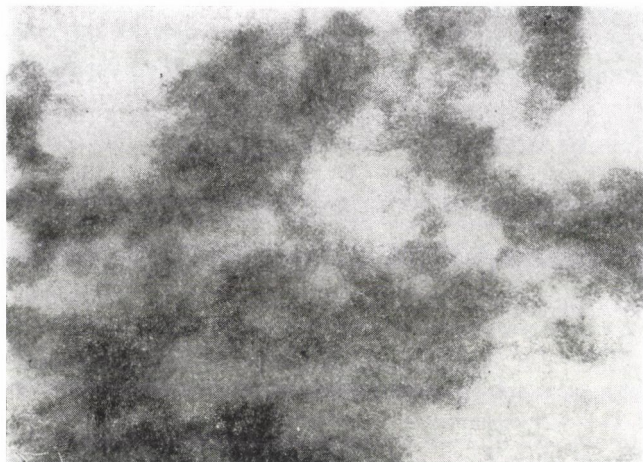


Fig. 5. Transmission electron micrograph of root tip myosin ( $\times 165\,000$ )

Ultra-thin sections of myosin filament aggregates were prepared by the Durcupan technique.

This myosin binds rabbit muscle actin, and the activity of ATPase is increased in the presence of  $Mg^{2+}$ .

Immunity testing of the root tip myosin is in progress.

### Acknowledgement

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### References

- BRITZ, S. J. (1979): Cytoplasmic streaming in *Physarum*. In Haupt, W.—Feinleib, M. E. (eds): *Encyclopedia of Plant Physiology*. Vol. 7. Springer, Berlin—Heidelberg—New York. 127–149.
- CLARKE, M.—SPUDICH, J. A. (1977): Nonmuscle contractile proteins: The role of actin and myosin in cell motility and shape determination. *Ann. Rev. Biochem.*, **46**, 797–822.
- FAZEKAS, S.—SZÉKESSY-HERMANN, V.—ÓVÁRY, I. (1979): Decrease and autophosphorylation increase of the labile phosphate content in myosin. *Acta Agron. Acad. Sci. Hung.*, **28**, 301–312.
- FAZEKAS, S.—SZÉKESSY-HERMANN, V.—ÓVÁRY, I.—KÁSA, I. (1980): Characterization of NaCl-extracted and purified myosin. *Acta Agron. Acad. Sci. Hung.*, **29**, 39–46.
- FAZEKAS, S.—SAMU, J.—SZABÓ, E.—SZÉKESSY-HERMANN, V. (1981): Identification and specific reactions of alkali stable amino acid phosphates in myosin hydrolysates. *Acta Agron. Acad. Sci. Hung.*, **30**, 340–350.
- FISKE, H. C.—SUBBAROW, Y. (1925): The colorimetric determination of phosphorus. *J. Biol. Chem.*, **66**, 375–400.
- FOLCH, J.—LEES, H.—SLOANE-STANLEY, G. H. (1957): A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.*, **226**, 197–209.

- GOA, J.—SCAN, D. J. (1963): A micro-biuret method for protein determination of total protein in cerebral fluids. *J. Clin. Lab. Invest.*, **2**, 218–223.
- JACKSON, W. T.—DOYLE, B. G. (1977): Characterization of actin of from root-tips of *Phaseolus vulgaris*. *J. Cell. Biol.*, **75**, 268a.
- KATO, T.—TONOMURA, Y. (1977): Identification of myosin in *Nitella flexilis*. *J. Biochem.*, **82**, 777–782.
- LOWRY, O. H.—LOPEZ, J. A. (1942): The determination of inorganic phosphate in presence of labile phosphate esters. *J. Biol. Chem.*, **162**, 421–428.
- METCALF III, T. N.—SZABO, L. J.—SCHUBERT, K. R.—WANG, J. L. (1980): Immunochemical identification of an actin-like protein from soybean seedlings. *Nature*, **28**, 5761, 171–172.
- NEHÉZ, R.—FAZEKAS, S.—ÓVÁRY, I.—SZÉKESSYÉ-HERMANN, V. (1980): Purification and some properties of myosin prepared from tips of root of *Zea mays* L. (hybrid SZE DC 384). *Proc. 20th Hung. Annu. Meet. Biochem.*, Siófok, 245–246.
- RUBIN, R. W.—COUSINS, E. H. (1976): Isolation of a tubulin-like protein from *Phaseolus*. *Phytochem.*, **15**, 1837–1839.
- VAKAY, M.—TRAUTWEIN, R.—SCORDILIS, S. P. (1978): Actin and myosin from the higher plant *Lycopersicum esculentum*. *Biophys. J.*, **21**, 23a.
- WAGNER, G. (1979): Actomyosin as a basic mechanism of movement in animals and plants. In Haupt, W.—Feinleib, M. E. (eds): *Encyclopedia of Plant Physiology*. Vol. 7. Springer, Berlin—Heidelberg—New York. 114–126.





## VARIA

### AUTUMNISATION\*

#### Introduction

In the case of wheat, the transformation of non-hardy spring wheat into hardy winter wheat is called autumnisation. For inducing autumnisation as a genetic variation adequate to a change in the environment (spring type → winter type), the spring sowings usual for spring wheat are replaced with properly timed autumn sowings.

Winter and spring habit, as a Mendelian character transmitted by gametes, is a hereditary property in several species of *Gramineae*, including wheat. Winter hardiness is associated with winter type, though the former is not exclusively a hereditary characteristic of winter wheat; winter hardy spring wheats, i.e. intermediate wheats also exist.

The universally acknowledged achievements of the "Mironovka" winter wheat breeding programme, based on autumnisation, and in particular the variety "Mironovskaya" 808, which was selected from an autumnised population of the spring wheat "Artemovka" and is cultivated over many millions of hectares, have caused MAKSIMCHUK (1963) to regard autumnisation as a breeding method of great promise, which deserves to be given adequate status in wheat breeding.

#### A short history of autumnisation

LINNAEUS (1753) classified spring and winter wheats as two distinct species: *T. aestivum* and *T. hybernum*. The concept of the permanence of species accepted by Linnaeus excluded the transformation of winter and spring wheats into each other as they were regarded as two different species. In 1778 LAMARCK (1795) re-classified winter and spring wheats as one species, *T. sativum*, thus reducing the differences assumed to exist between them under the Linnaean system.

In the 19th century the view arose that the difference between winter and spring wheat was insignificant, and that the forms could thus be easily transformed into each other. However, this was doubted long ago, first by KÖRNICKE (1885) and then by VAVILOV and KUZNETSOVA (1921), who did not regard the known experimental facts as hereditary transformation, but considered the "initial plant stock of such experiments as not being of pure lines or homozygotic with respect to vegetation period".

The anti-Darwinistic view that heredity cannot be changed under the effect of the environment, which became wide-spread among geneticists from the beginning of the 20th century, resulted in the almost total omission of the transformation of spring and winter

\* English version of a paper published in Hungarian in a book entitled "Wheat manual".

wheats into each other from research programmes. This explains why, when LYSENKO (1937), in the middle of the 1930s, re-posed the question of the transformation of spring and winter wheats into each other, the majority of geneticists treated the topic as a new problem. It is true, as SKRIPCHINSKY (1955) correctly pointed out, that both Lysenko and succeeding authors generally forgot to mention the names of their predecessors in publications on the subject.

Since the mid-1930s, the original and basic purpose of autumnisation experiments has been the clarification of the conditions necessary at a particular moment in the development of spring plants in order to change their heredity towards winter type (LYSENKO 1937). Since research was begun on the subject almost half-a-century ago, about 200 papers have been published on autumnisation, clarifying several aspects of the process.

In the "sowing times" method generally used in autumnisation, the first autumn sowing — as LYSENKO (1952, 1963) showed — serves the purpose of destabilising the spring type of heredity, or spring habit, while the second autumn sowing is aimed at the development of the winter type of heredity, or winter habit. Two properly chosen autumn croppings proved to be sufficient for autumnisation in the experiments of certain authors. Other authors hold that in general more than two autumn croppings are necessary for autumnisation. According to the reports published by a few authors, in certain cases one autumn cropping was sufficient to achieve autumnisation. All the authors suggest that the second autumn sowing be carried out close to the optimum sowing time winter wheat. Some of the authors advocate that the first year's sowing be done late, in early winter, while others advise an early autumn sowing. KOZLOVA's (1956) statement that different spring varieties may require different conditions for autumnisation even at the same experimental site deserves attention.

The winter hardiness and frost resistance of winter forms obtained through autumnisation may be as great as those of the winter wheat varieties native to the particular region (LUKYANENKO 1948). The erect growth habit of the initial spring varieties changes to the prostrate growth habit typical of winter wheats. Autumn growth slows down. The hairiness of the first leaf disappears. The greyish-green leaf colour is supplanted by a dark-green colour in certain varieties. Varietas changes may occur, etc.

In the autumnisation experiments of GLINYANY (1963) a larger number of autumnised plants were derived from the progeny of the main spikes of the primary shoots, formed from apices developing in early autumn, than from the progeny of the subsidiary spikes of the secondary shoots, formed on the same plants from apices appearing later in the autumn. This is a case of tillers developing under different environmental conditions on the same plant and of the assumed genetic consequences of this phenomenon. On this basis the heterogeneities noted in the vegetation period and in other characteristics in the autumnisation population can be justly interpreted as adequate genetic variations. The studies of TRUKHINOVA (1950), O. LYSENKO (1956) and others proved that the winter forms arising from spring wheats in various climatic zones are generally adjusted to the climatic conditions of the particular zones, i.e. they are steppe, forest-steppe, etc. ecotypes. Yet, the resistance to the rigours of the weather in a given region, arising as an adaptation of the developing winter forms obtained through autumnisation, can never be absolute because the autumn weather conditions are not the same in different years in one and the same region. Therefore, the winter forms developing in the same region under different autumn conditions in different years may differ from one another. These factors, both together and separately, provide a splendid opportunity for the assertion of natural selection among the autumnised plants of the autumnisation populations.

On the basis of experimentation on the transformation of spring and winter wheats into each other, LYSENKO (1963) formulated one of the fundamental rules governing the life and development of living creatures, the law of adequacy. According to this law, any change



in the heredity of an organism is adequate; that is, it corresponds to the influence of the changed environmental conditions which it has assimilated. Autumnisation as an adequate genetic variation cannot be satisfactorily interpreted by a genetic concept which denies the inheritance of acquired characters. This for the most part explains the objections to be found in genetic literature to the positive facts of the autumnisation experiments. These objections, almost without exception, are, like those of VAVILOV and KUZNETSOVA (1921), concerned with the incorrect choice and control of the initial plant stock and certain related errors in methodology.

We can only agree with SKRIPCHINSKY (1935) when he stipulates the accurate choice and control of the experimental plant material in order to ensure its freedom from mechanical and biological contamination, i.e. that there be no winter forms or spring  $\times$  winter hybrids already present in the spring wheat plant stock used for autumnisation experiments. In strict pedigree selection on a single plant basis (line  $\rightarrow$  sub-lines, etc.) the methodology should ensure the precise control of the spring and winter types, so that any plant determined as being of winter or spring type should be really so genetically. Every spike whose grains will later be used for sowing must be isolated during the autumnisation experiments. For similar reasons Skripchinsky requires the phenological observation of every single plant, e.g. the separate recording of the heading of each plant. It can be seen from the autumnisation literature that no autumnisation experiment fully met the stipulations regarding the choice of experimental material and methodology rightfully raised by Skripchinsky, or at most only from certain aspects.

Again we must agree with SKRIPCHINSKY (1957) who, in the words of DARWIN (1868), gives a categoric "No" to those who "imagine that natural selection induces variability" and expressively emphasizes that selection "implies only the preservation of such variations as arise and are beneficial to the being under the conditions of life".

#### Autumnisation at Martonvásár

##### *a) In the field*

Simultaneously with the first crosses in the winter wheat breeding programme, which started from scratch, genetic and physiological investigations on winter habit and winter hardiness began in autumn 1955. Autumnisation, the study of the external and internal conditions required for the transformation of non-hardy spring wheat into hardy winter wheat, was chosen as a general method for describing these characters and determining the conditions under which they develop. But since the very possibility of autumnisation was in general the subject of debate, the experimentation began with tests to show whether autumnisation existed. The starting point for the autumnisation experiments was the fact that, in the initial stage of development, spring wheat does not require a temperature around the freezing point or shortening daylength in order to flower. The sowing times method (ecological sowings) employed in the autumn proved, however, that the genetic constitution of spring wheat can be changed to such an extent that exposure to temperatures near the freezing point and shortening daylength are essential if flowering is to be induced, i.e. spring wheat can be autumnised. Depending on cycles or years and, to some extent, on the initial varieties, 2, 3, 4, 11, 13, 14 or 15 autumn croppings were required for the induction of autumnisation in field experiments which fully complied with the requirements raised concerning the selection of initial plant stock and methodology (SKRIPCHINSKY 1955, 1957). Spring-sown progeny tests, conventional and aneuploid genetic analyses, combined with multiple physiological tests (vernalisation, photoperiod, winter hardiness, chlorophyll content, assimilation temperature, organic matter accumulation, intensity of growth and cell division, enzyme activity) all con-



firmed that the initial stock of the Martonvásár experiments was non-hardy spring wheat, while the autumnised variants were hardy winter wheat (RAJKI 1967, 1982; RAJKI and RAJKI 1969).

The most detailed and severe criticism of the academic doctoral thesis on autumnisation (RAJKI 1967) was published in "Botanichesky Zhurnal" (AGAEV 1969). In this criticism, which rejected the genetic interpretation of autumnisation given in the thesis, the most positive aspects of the experimentation were summarised in the following way: "It can be seen from Rajki's experiments that the development of new forms can be directed to a large extent by the method of ecological sowings (in autumnisation experiments). If the autumn generations of spring wheat are interrupted before the appearance of intermediate forms by a single spring generation, no intermediate forms will later appear among the experimental spring wheat plants, however many generations are sown in the autumn. In an analogous manner, it is possible to stop the process of transformation from spring to winter plants in the intermediate or semi-winter phase. Obviously, this discovery makes it possible to analyse the laws governing the development of new forms through the autumnisation process much more profoundly than before, and it is also one of the most convincing proofs of the fact that a hereditary transformation of spring plants into winter forms really occurs during the autumnisation experiments." In the journal "Genetika" these merits were complemented with the statement that the higher the standard of experimentation, the more pronounced the appearance of intermediate wheat as a connecting, indispensable "medial" link in the autumnisation process (KRIVOGORNITSYN 1978).

#### b) *In the phytotron*

At the end of the fifties, when the first cases of autumnisation were being interpreted, it was decided to study those aspects of the transformation into hardy winter wheat which were a function of the variety or of the environmental conditions (particularly the meteorological conditions), which were themselves determined by the sowing time. Several years of examination made it clear, however, that without modern plant raising and testing chambers the difficulties faced in determining correlations of this type were insurmountable. This was the fundamental motivation for the establishment of the Martonvásár phytotron, and consequently for the development and continual improvement of plant raising methods which simulate nature and are unique in the history of phytotrons (DOWNS and HELLMERS 1976). The trends in air temperature, humidity and illumination values are calculated from data provided by the Martonvásár Agrometeorological Observatory (PLETSEY 1973) using trigonometrical curve fitting. When elaborating the climatic programmes, it is assumed that the weather remains constant for seven days. In order to simulate nature as faithfully as possible, daily temperature rhythms appropriate to the season are used instead of the traditional stable day and night temperatures, and a daily rhythm for the duration and intensity of illumination, altering with the change in the seasons, is programmed instead of the usual photoperiods. The climatic programmes, particularly the autumn ones (marked A plus a serial number, e.g. A24) are very diverse. The autumn climatic programmes include some based on the most frequent air temperature, air humidity and illumination averages starting from the middle of September, the end of October, or the beginning of November, etc. But far more of the autumn programmes are based on the meteorological parameters of cooler but sunnier years, which occur in nature less often, and in some cases these are corrected for special experimental purposes. The spring climatic programmes (marked S plus serial number), like the autumn ones, also simulate nature. The faithfulness of the simulation was checked over a period of several years by carrying out field experiments combined with parallel experiments in the phytotron, where the field conditions were simulated with a one-week delay. The only

exception to the principle of simulating nature is the winter programme (W plus serial number), where, except in the special frost resistance tests, temperatures around 0 °C are generally programmed. And plants grown on these climatic programmes are stocky, strongly tillering plants with the growth type, culm length, straw stiffness, productive tillering and ear productivity characteristic of the variety; in other words, they are in no way deficient compared to winter wheat plants grown under optimum field conditions. A great advantage of the climatic programmes is that they allow the field vegetation period of winter wheat to be reduced to approximately half in the phytotron. This was achieved because, understandably, the stress effects occurring in nature (temporary drought, extreme fluctuations in temperature, etc.) were not programmed in the phytotron except when it was the consequences of the stress effects which were to be studied.

In the first five or six years of experimentation, the repeated use of a large number of climatic programmes, elaborated after an analysis of the climatic conditions in the field during the autumnisation of classical and Mexican spring wheats, led, reproducibly, to a maximum two-week delay in the heading of the spring wheats examined. This is quite literally a half-success, since under Hungarian conditions autumnisation cannot be registered unless there is a delay in heading of at least one month.

In order to develop the climatic programmes required for reproducible autumnisation, a thorough review of the previous attempts at field and phytotron autumnisation was begun, as so often before, in order to decide why success was only partial. Food for meditation was given by the fact that in the second cycle of autumnisation experiments, which began in 1957, in two variants (a spring sowing was included after the first two autumn sowings in the first variant and after the first three autumn sowings in the second, with the result that the first behaved as spring wheat and the second as intermediate wheat) autumnisation was recorded in the early seventies, viz. after the 15th (16-1) autumn sowing for the spring variant, and after the 14th (15 - 1) autumn sowing for the intermediate variant, i.e. in the two variants which had already been mentioned in the criticism published by AGAEV (1969) and in the statement made by KRIVOGORNITSYN (1978). When seeking for the causes of this phenomenon in the presumed relationship between the delayed autumnisation as compared to the usual, and the autumn and winter meteorological conditions of the years preceding the phenomenon, it was found that the winter of 1969/70, a year or two before the stubborn spring and intermediate variants were finally autumnised, was exceptional for the large number of days with snow cover (snow depth > 1 cm). There were 92 days with snow cover in 1969/70, compared with an average of 33 days for the 5 previous winters (extreme values 20 and 39). Beneath the snow cover there is generally a temperature of around 0 °C, which is active for vernalisation, and this may favour the development of winter habit as an acquired character. The total autumn/winter period with a temperature active for vernalisation is obviously made up of the days with snow cover, combined with the days and hours when the temperature is around 0 °C despite the lack of snow cover. From year to year there is a certain difference in the ratio of days with snow cover to those with no snow cover but with a temperature active for vernalisation, and this influences the light conditions, which are also important for autumnisation. On the assumption that days with snow cover, or the microclimate under the snow cover, are important for autumnisation, the testing of winter climatic programmes lasting 12 or even 18 weeks was introduced in addition to the earlier 6-7-week winter. At the beginning of the eighties, two of these long winter treatments produced the first autumnisations in Penjamo 62 (RAJKI 1982).

In one of these treatments, 12- or 18-week winters were programmed for some sublines in the third generation, after two winter programmes of normal, i.e. 6-week length. The 12-week winter programme did not produce anything new, but autumnisation was recorded in spring-sown progeny tests on plants raised on the 18-week winter programme (A26 + W9 +



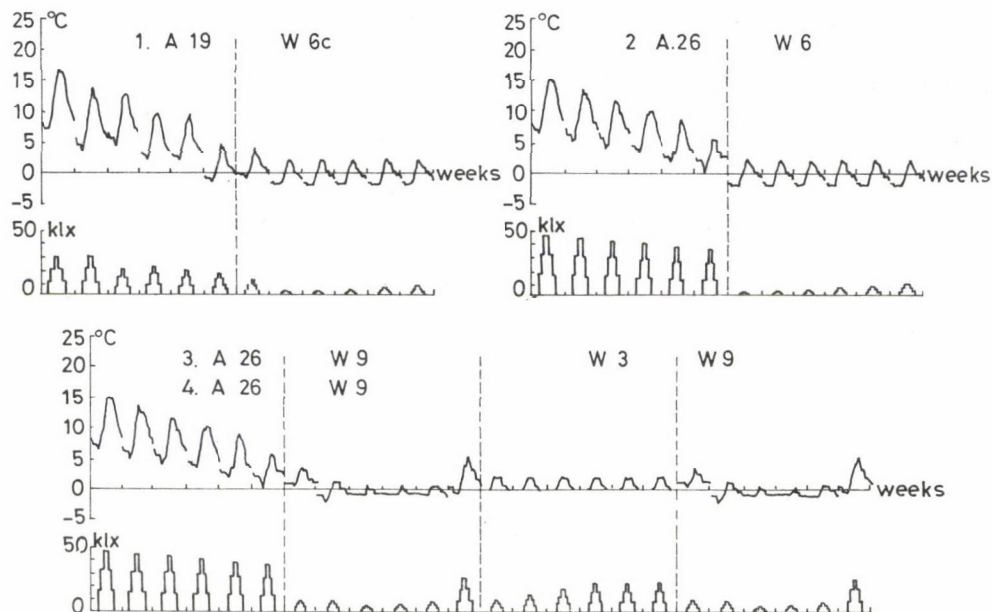


Fig. 1. Climatic programmes for one of the successful autumnisation experiments in the phytotron

W3 + W9; Fig. 1). After the subsequent, fourth generation, where a 6-week winter was programmed, the autumnisation was confirmed. A similar picture was found if a slightly different 18-week winter programme was used in the third autumnisation generation.

In the second case, 12- and 18-week winters were not programmed until the fifth autumnisation generation. Apart from the difference in the number of generations, the structure of the two successful autumnisation treatments only differed in the autumn programme of the first generations. The A19 programme used in the first case was modified in such a way (programme A9) that every other week the temperature was 1–2 °C higher and the illumination approximately 10% weaker, but naturally these, too, simulate autumn weather conditions.

In the spring-sown progeny test on the fifth autumnisation generation, some of the plants did not head, i.e. they were autumnised. Using the grain yield of plants which headed with an average delay of 17 days in the spring-sown progeny test, the experiments were continued in two directions: a) Some was used for a further progeny test in the field, which confirmed the results of the phytotron progeny test: of the 5 sub-lines, 2 did not head, while the other 3 only partially (27–33%) headed. b) At the same time vernalised seedlings were planted in the field for propagation purposes. In a further spring-sown progeny test, using some of the grain yield from the propagation, 11 of the 15 sub-lines did not head, while 4 headed only partially and/or with a delay in heading. The remainder of the grain yield obtained from the propagation, and seed from autumnisation generations which preceded the long winter treatment, were used for frost resistance testing in the phytotron. The frost test was developed at Martonvásár during the seventies (RAJKI 1980). The preliminary raising programmes, which prepare the seedlings for freezing, simulate nature and are thus an original solution. Without climatic programmes which promote normal growth and development it would be virtually impossible to reliably and consistently test the frost resistance of the



experimental plants independently of annual fluctuations in the weather. As regards the results of the phytotron test (Table 1), the frost resistance of the best sub-lines of the autumnisation population proved no worse than that of the standard winter wheat. However, the frost resistance both of autumnisation generations which preceded the long winter treatment and of the initial spring variety gave a value of 0 (RAJKI 1982).

Table 1

*Survival of autumnising and autumnised lines of the spring wheat Penjamo 62 frozen at  $-15^{\circ}\text{C}$*

No.	Variants	Survival %
1.	Penjamo 62	0
2.	(A9 + W6c) + (A26 + W3)	0
3.	(A9 + W6c) + (A26 + W3) + (A24 + W6)	0
4.	(A9 + W6c) + (A26 + W6) + (A26 + W3)	0
5.	(A9 + W6c) + (A26 + W6) + (A26 + W3) + (A26 + W9) + + (A24 + W6 + 3 + 9)	76.3
6.	(A9 + W6c) + (A26 + W6) + (A26 + W3) + (A26 + W9) + + (A24 + W6 + 3 + 9) + [SPTP]*	87.5
7.	(A9 + W6c) + (A26 + W6) + (A26 + W3) + (A26 + W9) + + (A24 + W6 + 3 + 9) + [SPTP] + [SSVS]**	87.3
8.	Libellula: slightly hardy Italian winter wheat	7.5
9.	Martonvásár 4: standard winter wheat in Hungary	79.8
10.	Mironovskaya 808: very hardy Russian winter wheat	92.5

\* [SPTP] = Spring Progeny Test in the Phytotron, i.e. grains from the 11 plants which headed in the progeny test of the 5th autumnisation generation.

\*\* [SSVS] = Spring Sown Vernalised Seeds, i.e. grains from plants of a field propagation sown with vernalised [SPTP] seeds in spring.

Despite the promising results obtained in phytotron autumnisation, new problems have also arisen. While the spring wheat plants survive the 6-week winter programmes fairly well, there are difficulties in plant raising during long winters. In contrast to winter wheat, organic matter accumulation virtually ceases, cell division and growth are retarded, while tillering and the differentiation of the growing tip continue in spring wheat as the temperature approaches the freezing point (RAJKI 1967). Differentiation may reach stage II, III or even IV on the KUPERMAN (1953) scale during the phytotronic winter, depending on the variety. However, vernalisation is completed during stage III, after which it is no longer possible to influence the process. Consequently, during the phytotronic winter, a few hours of temperatures just above the freezing point are programmed to ensure the water supply, interspersed with periods just below the freezing point in order to slow down the differentiation of the growing tip. But the winters in the phytotron are more severe than in the field, because no sooner has the temperature in the chamber sunk below zero when the soil in the pots, together with the entire root system, freezes. In order to avoid overdevelopment of the plants, above-zero temperatures cannot be programmed for more than a few hours a day, which is not sufficient to thaw out the frozen root system. At the same time, to however minimal an extent, the above-ground organs of the plant are renewing their physiological activity, but the water supply from the frozen soil and root system is uncertain, causing the

plants to suffer and in some cases to die. Several solutions to the problem are conceivable, among which the further modification of the climatic programmes, soil conditioning independent of the air conditioning, and the use of trickle irrigation were in progress, with a fair chance of success (RAJKI 1982), when the autumnisation at Martonvásár came to an end in 1983.

### Autumnisation and the dilemma of genetics

The results of the autumnisation experiments have been published in about fifty scientific papers and at least as many lectures, delivered mostly in Western Europe and North America. The most recent lecture (RAJKI 1982) was presented in August 1981 at the invitation of the World Congress of Botany (Sydney University, Australia). The Sydney lecture created quite a stir in New Zealand, Japan and China, too, and was repeated in the form of a seminar arranged by the Academia Sinica in September 1981 in Peking, Nanking and Shanghai. The intriguing discussions which followed the lectures, made it possible to give a comprehensive analysis of the practical as well as the theoretical aspects of autumnisation. The plant breeding aspects of autumnisation can hardly be disputed. The situation is quite different for the genetic aspects of autumnisation: the lack of comprehension, often amounting to a stubborn rejection, has been quite unaffected by the efforts of STEELE (1979), who interprets the inheritance of acquired immunological tolerance as a Lamarckian phenomenon. As was to be expected on the basis of earlier experience, it proved necessary, time and again, to refute WEISMANN's (1891-1892) experiment on mice, designed as a denial of the Darwinian pangenesis hypothesis, which was conceived in the spirit of the Lamarckian phenomenon: the tail cannot be amputated until it is formed, i.e. before the hypothetical gemmules have reached the gametes from the tail. Of course, the germ plasm theory of molecular genetics, the central dogma (CRICK 1958, 1970), which postulates the impossibility of any reversed or unknown transfer of information, stood in the focus of the discussions, since this doctrine excludes the possibility of an adequate variation. But, as Crick notes, there is absolutely no proof for the central dogma, and by its very name he tried to emphasize its speculative nature. The almost generally accepted view in genetics that viruses and other specific substances cannot induce specific resistance in the form of a genetic variation taking place in the organism, i.e. new genes can only be produced by accident, is fundamentally nurtured by the indirect proof obtained from a single experiment, the LURIA and DELBRÜCK (1943) fluctuation test. However, the probative force of the fluctuation analysis as a historical parallel of the Weismann's test is marred by the same sophism as the experiment on mice with amputated tails: virus resistance cannot be tested if the virus is not present.

As a matter of fact the central dogma, being "a negative statement" (CRICK 1970), is undemonstrable, but according to the rules of logic its contrary is to be proved. In the same way the proof of the existence of the gene is a logical absurdity, because according to the classical and/or molecular gene concept this not only signifies the genetic material, but also expresses the specific relationship between heredity and metabolism, the body and its environment, i.e. the impossibility of any adequate transcription of the information due to changes in the environment or the metabolism. Here, too, only the contrary can be proved. It is for this reason that the irrefutable proof of any adequate genetic variation, or to quote CRICK (1970), the realisation of any of the "unknown" transfers, would "shake the whole intellectual basis of molecular biology". Similarly, it is only "judicious" to discourse on the preparation of a gene, i.e. the genetic material independent of changes in the environment or the metabolism, as long as no adequate variation has been irrefutably proved.

Despite the weakness of the experimental results opposing the Lamarckian phenomenon and the more and more convincing experimental proofs supporting this phenomenon,



I do not believe that "the orthodox doctrines . . . are crumbling anyway, and in a decade or two will have vanished into limbo, as other orthodoxies have in the past" (KOESTLER 1980). Because to rearrange a well-known series of data, to judge it differently and to avoid the prevalent doctrine is the most difficult of intellectual acts. This is a practically invincible intellectual barrier, which is to be found beside the cradle of every scientific discovery. But however discouraging, or even injurious, the resistance to new discoveries may be, it does have a certain value in that it protects science from the rash acceptance of ideas which are not sufficiently proved or tested. Nothing can cause greater damage to science than the abandoning of a critical standpoint and the easy acceptance of hypotheses supported by incomplete and half-tested proofs. However, a critical standpoint is by no means identical with mere scepticism or rigid rejection. Scientific progress is inconceivable without a thorough investigation of anomalies and without the freedom to express and defend minority opinions.

The prospects — as I see them — are not rosy at all: over the last half-century science has changed for the worse and this tendency seems to be continuing. The world of science has become deformed (CHARGAFF 1978), especially since the second World War. Research, the cultivation of science, has become a job — a situation which was previously quite unknown. Years ago science was the preserve of scholarly professors whose principal vocation was teaching. The study of science is now a mass occupation, which is a concomitant of the sudden, unhealthy increase in science in a world which has lost all sense of proportion, mainly because there are many who regard a scientific post as a sinecure. At the same time the administration of science has also become a new category of job, which was again previously unknown, since the font of science, the university, was hardly administered at all, luckily for our predecessors.

There was a time when no-one would have dreamed of asking what the point of research was: it was considered a good thing to learn more about the world we live in. It did not occur to anyone to ask for an official definition of the research aim or for a research programme. In the meantime the idea of biology as a devoted, modest, conscientious examination of life has given way to the clamorous, unscrupulous pursuit of "break-throughs", for the ever greater financial support of more and more costly research and for a better researcher's salary. If the present "programme orientation" and profit-seeking had existed in the past, the majority of great scientists would never have emerged: in fact, science itself might never have been born.

The professional researcher is generally obsessed with a single problem in a specialised field. The problem may be highly specialised in that it concerns only a small fraction of the discipline in question (in this case, biology) and/or in that it deals with only a tiny portion of the organism under examination. As a consequence, the results of the experiments are of an extremely particular nature, which is reflected in their value, since they are only valid for a fragment of the discipline or organism, and not for the whole. The hope that these fragments of knowledge will one day fit together to give a congruent whole has so far not come true, nor is it likely to in the future, since the more we differentiate the more difficult it becomes to integrate.

Years ago a biologist carrying out original research was generally at the heart of the discipline, but inordinate specialisation has pushed almost everyone to the periphery. During the last 50 years, as in the past, the heart of biology has been the cardinal question of the Lamarckian phenomenon or its modern equivalent, adequate genetic variation, which invalidate Weismann's germ plasm theory and its modern counterpart. Crick's central dogma. It is because of this dilemma, or of the situation which has established itself in genetics, that WADDINGTON (1968), who is described by MAYNARD-SMITH (1971) as "having performed perhaps the most illuminating anti-Lamarckian experiment ever devised", wrote: "For reasons which it would be invidious to go into in any historical detail, Neo-Darwinism has become an established orthodoxy, any criticism of which is regarded as little less than *lèse-majesté*."



But what is the *modus vivendi* to be? For anyone meditating on a solution to the problem it is far from comforting to know that there is no sign as yet of a panacea being found for mankind's numerous other ills either, such as the present and future deficiencies in the supplies of healthy food, pure water and unpolluted air, or the unimaginable and ever more ominous contamination of the human mind and body and of the natural and mental environment. Looking back over the years, I think perhaps my greatest scientific error was when I assumed in the mid-sixties that the best way to understand and control heredity was by a thorough and manifold understanding of the biochemistry of metabolism (RAJKI 1967). My meditations over the past 15 years and the thoughts of others (LYSENKO, CHARGAFF, KOESTLER), which confirmed the modifications in my own thinking, finally led me to the conclusion that, when making this hypothesis, I fell into the trap of reductionism. After all, the result of a biochemical analysis — however necessary and essential it may be — is no more than just another type of description and is not the crux of the phenomenon of life. Especially if we consider that "life is what is lost in the test-tube" (CHARGAFF 1963). And even if the absurdly reductionist statement that Man could be ultimately defined as "nothing but 90% water and 10% minerals" were correct, where does that get us? (KOESTLER 1980). So I can only agree with the prophecy that the greatest of all revolutions is still to come. one which will liberate science from the fetters of mechanistic thinking (CHARGAFF 1978).

#### And what about wheat breeding?

The above-cited, positive evaluation made by MAKSIMCHUK (1963) — certainly a pragmatist as a plant breeder — on autumnisation as a method of obtaining a breeding stock for selection, is valid irrespective of the genetic controversy, so, why should autumnisation as such not be applied in practical plant breeding? Or is the matter perhaps not so simple after all? Despite the fact that we possess an autumnised version of the best Mexican spring wheat, Siete Cerros, with a fifty-day vernalisation requirement and reasonably good frost resistance?

If autumnisation is treated not as a genetic but as a breeding method the need for a strict pedigree (line → sub-lines, etc.) and the manifold checking procedures can be eliminated. A few ecological sowings around the optimum sowing date for winter wheat in the first and second autumn croppings of an initial spring wheat variety — if carefully chosen for the breeding purposes — could lead to the development of an excellent autumnised plant stock. And here successful selection could be of immediate practical value (REMESLO 1970).

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#### References

- AGAEV, M. G. (1969): K probleme eksperimental'nogo vzaimoprevrashcheniya yarovykh i ozimnykh rastenii. Nekotorye soobrazheniya v svyazi s issledovaniyami Sandor Rajki po ozimizatsii (On the problem of transformation of spring plants into winter plants and vice versa. Certain considerations in connection with the investigations of Sandor Rajki on autumnisation). *Bot. zhurnal*, **54**, 1364–1378.
- CHARGAFF, E. (1963): *Essays on Nucleic Acids*. Elsevier Publishing Co. Amsterdam, London, New York, 212.
- CHARGAFF, E. (1978): *Heraclitean Fire*. The Rockefeller University Press, New York, 252.
- CRICK, F. H. C. (1958): On protein synthesis. *Symp. Soc. Exp. Biol.*, **12**, 138–163.
- CRICK, F. H. C. (1970): Central dogma of molecular biology. *Nature*, **227**, 561–563.
- DARWIN, CH. (1868): *The Variation of Animals and Plants under Domestication*. John Murray, London, 482.

- DOWNES, R. J. and HELLMERS, H. (1976): Controlled Climate and Plant Research. Technical Note 148. World Meteorological Org. Geneva, 60.
- GLINYANY, N. P. (1963): Rezultaty izucheniya processa prevrashcheniya yarovykh rasteniy v ozimye (Results of studying the process of the conversion of spring into winter plants). In: Upravlenie nasledstvennostyu sel'skokhozyaystvennykh rasteniy. Izd-vo s/kh lit. Moskva, 122-126.
- KOESTLER, A. (1980): Bricks to Babel. Hutchinson, London etc. 697.
- KOZLOVA, A. (1956): Opyt peredelki yarovoy pshenitsy v ozimuyu (Experiment on the conversion of spring into winter wheat). *Agrobiologiya*, **3**, 65-68.
- KÖRNICKE, F. (1885): Die Arten und Varietäten des Getreides. In: Handbuch des Getreidebaues, I. Parey, Berlin, 6 + 470 + X.
- KRIVOGORNITSYN, B. I. (1978): K voprosu vzaimoprevrashcheniya yarovoy i ozimoy pshenitsy (On the problem of transformation of spring into winter wheat and vice versa). *Genetika*, **4**, 536-544.
- KUPERMAN, F. M. (1953): Biologicheskie osnovy kul'tury pshenitsy (Biological bases of wheat cultivation). Izd-vo Moskovskogo Univerziteta, Moskva, 300.
- LAMARCK, J. B. P. (1795): Flore française, I-III. Paris, 1740.
- LINNAEUS, C. (1753): Species Plantarum, I-II. Holmiae, 1200.
- LYSENKO, O. T. (1956): Klimaticheskie usloviya i osobennosti ozimyykh form, vznikshikh iz yarovykh (Climatic conditions and peculiarities of winter forms obtained from spring ones). *Agrobiologiya*, **3**, 71-77.
- LYSENKO, T. D. (1937): O dvukh napravleniyakh v genetike (On the two trends in genetics). *Yarovizatsiya*, **1**, 29-75.
- LYSENKO, T. D. (1952): *Agrobiologiya* (Agrobiology). Gos. izd-vo s/kh lit., Moskva, 781.
- LYSENKO, T. D. (1963): Teoreticheskie osnovy napravlennoy izmeneniya nasledstvennosti sel'skokhozyaystvennykh rastenii (Theoretical bases of the controlled change in the heredity of cultivated plants). In: Upravlenie nasledstvennostyu sel'skokhozyaystvennykh rasteniy. Izd-vo s/kh lit. Moskva, 7-25.
- LUKYANENKO, P. P. (1948): Izmenenie prirody sortov ozimoi i yarovoi pshenitsy putem izmeneniya uslovii prokhozheniya stadii yarovizatsii (Changing the nature of winter and spring wheat varieties by means of changing the conditions during the course of the vernalisation stage). *Agrobiologiya*, **2**, 40-50.
- LURIA, S. E. and DELBRÜCK, M. (1943): Mutations of bacteria from virus sensitivity to virus resistance. *Genetics*, **28**, 491-511.
- MAKSIMCHUK, L. P. (1963): Izmenenie porodnykh svoystv zernovykh i zernobobovykh kul'tur pod vliyaniem uslovii vyrashchivaniya i ispol'zovanie poluchaemogo materiala v selektsii (Changes in the hereditary traits of cereals and leguminous plants under the influence of raising conditions and the use of the material obtained in breeding). In: Upravlenie nasledstvennostyu sel'skokhozyaystvennykh rastenii. Izd-vo s/kh lit. Moskva, 213-219.
- MAYNARD-SMITH, J. (1971): The Case of the Midwife Toad by Arthur Koestler. *New Scientist*, **52**, 281-282.
- PLETSER, J. (1973): Climatic model for phytotron studies. *Acta Agron. Hung.*, **22**, 67-80.
- RAJKI, ERNA (1980): Testing for Cold Resistance in Cereals. FAO, Rome, 92.
- RAJKI, ERNA and RAJKI, S. (1969): Monosomic analysis of growth habit in the autumnisation process. Fifth Congress of the Eucarpia, Milan 1968. *Genetica Agraria*, 43-47.
- RAJKI, S. (1967): Autumnisation and its Genetic Interpretation. *Akadémiai Kiadó, Budapest*, 88.
- RAJKI, S. (1982): Phytotron in the service of genetics and wheat breeding. *Acta Agron. Hung.*, **31**, 421-434.
- REMESLO, V. N. (1970): Nekotorye itogi selektsii ozimoi pshenitsy (Some results of breeding winter wheat). *S/kh biol.*, **5**, 197-206.
- STEELE, E. J. (1979): Somatic Selection and Adaptive Evolution. On the Inheritance of Acquired Characters. Williams-Wallace International Inc. Toronto—Croom Helm Ltd. London, 90.
- SKRIPCHINSKY, V. V. (1955): Prevrashchenie ozimyykh zlakov v yarovye i yarovykh v ozimye v svete ucheniya Darvina (Conversion of winter into spring cereals and of spring into winter cereals in the light of the doctrine of Darwin). *Bot. zhurnal*, **40**, 64-90.
- SKRIPCHINSKY, V. V. (1957): Eshcho raz o prevrashchenii ozimyykh zlakov v yarovye i yarovykh v ozimye v svete ucheniya Darvina (Once more on the conversion of winter into spring cereals and of spring into winter cereals in the light of the doctrine of Darwin). *Bot. zhurnal*, **42**, 610-624.
- TRUKHINOVA, A. T. (1950): Napravlennoe izmenenie yarovoi pshenitsy Milturum 321 v ozi-



- muyu v usloviyakh Sibiri i Yuzhnogo Urala (Controlled change of the spring wheat Milturum 321 into winter wheat under the conditions of Siberia and South-Ural). Trudy inst. gen., **13**, 66-99.
- VAVILOV, N. I. and KUZNETSOVA, E. S. (1921): O geneticheskoi prirode ozimyykh i yarovyykh rastenii (On the genetic nature of winter and spring plants). Izv. Agr. Fak. Saratovskogo Universiteta, **1**, 1-25.
- WADDINGTON, C. H. (1968): Towards a theoretical biology. *Nature*, **218**, 1211-1213.
- WEISMANN, A. (1891-1892): Essays on Heredity, I-II. Oxford University Press, London, 471, 226.

#### HETEROSIS AND COMBINING ABILITY IN BARLEY

A diallel set of crosses, excluding reciprocals, involving ten parental lines of barley viz., PL 26, RD 47, K 13, GRA, WPG 52528, HB 16, EC 70849 and EB 4145 was used in this investigation. Parents and the 45  $F_1$  populations were grown in a randomized block design with three replications during 1977-78 at the Research Farm, Raja Balwant Singh College, Bichpuri, Agra. Each treatment was sown in one row plots of 5 meter length. The rows and plants within a row were spaced 25 cm and 15 cm apart respectively. Observations were recorded on five randomly selected plants from each plot on days to 70% heading, days to maturity, plant height, tillers per plant, leaf area, spikelets per spike, ear length, grains per ear, 250-grain weight and grain yield. Combining ability analysis was carried out following GRIFFING (1956b) Method II and Model I. Heterosis measured as deviation of the  $F_1$  mean from better parent was computed for selected hybrids.

The analysis of variance showed significant differences among the parents, hybrids and parents vs hybrids for all the characters (Table 1). It indicated that sufficient variability existed in the parents and their hybrids which accelerates the chances of new recombinations that can be isolated in the succeeding generations. The mean squares due to both general and specific combining ability for all the characters except sca variance for tillers per plant, were also highly significant (Table 2), indicating thereby the importance of both additive and non-additive variances. The magnitude of gca variances was however, larger than their respective s.c.a. variances for all the characters except spikelets per spike (0.873 : 1) and grains per ear (0.772 : 1). It revealed the predominant role of additive gene action for most of the characters except spikelets per spike and grains per ear where non-additive gene action was prevalent. KATARKI (1963), SMITH and LAMBERT (1968), TANDON *et al.* (1968), GULATI *et al.* (1969) and SOLANKI and BAKSHI (1974) have also reported high g.c.a./s.c.a. ratio for most of the characters in barley. In such cases where both the additive and non-additive genetic variances are present, superior genotypes may be obtained by adopting reciprocal recurrent selection. After a couple of cycles of recurrent selection the selected elite lines should be subjected to multilocation testing for further evaluation (COMSTOCK, ROBINSON and HARVEY, 1949).

General combining ability effects of the parents (Table 3) revealed that for days to maturity, UK 24, PL 28, PL 26, RD 47, K 13 and GRA were good cominers as they showed highly significant and negative g.c.a. effects for these traits. Out of these, UK 24, BD 27 and GRA exhibited consistent negative and significant g.c.a. effects for plant height also. For tillers per plant, UK 24 and RD 47 appeared to be good combining parents. The parents with desirable effects were PL 26, K 13, WPG 62528 and HB 16 for leaf area; HBL 6 for spikelets per spike and grains per ear and all the parents except WPG 62528, EC 70849 and EB 4145 for 250-grain weight. PL 28 was the best general combiner for ear length. RD 47 and GRA were the only parents which showed significant g.c.a. effects for grain yield. Overall, RD 47 and GRA appeared to be the best general combiners for days to 70% heading, days to maturity, plant height and grain yield, while PL 28 and HBL 6 were good combiners for spikelets per spike, grains per ear and 250-grain weight and therefore, use of these inbreds for developing early maturing and high yielding types would be desirable.



The estimates of specific combining ability effects are presented in Table 4. The cross combination UK 24  $\times$  PL 28 showed significant negative s.c.a. effect for days to 70% heading, while for days to maturity the cross PL 24  $\times$  RD 47 registered the highest negative value. For plant height the crosses UK 24  $\times$  EC 70849 and PL 28  $\times$  EC 70849 gave positive and significant effects. The highest negative value of 15.668 was recorded for the cross EC 70849  $\times$  EB 4145 which is likely to give dwarf segregants in future generations. Three crosses, UK 24  $\times$  RD 47, PL 26  $\times$  K 13 and GRA  $\times$  EC 70849 showed desirable s.c.a. effects for tillers per plant, whereas none of the crosses exhibited significant effect for leaf area. The crosses UK 24  $\times$  WPG 62528, PL 28  $\times$  WPG 62528, PL 28  $\times$  EC 70849 and GRA  $\times$  WPG 62528 for ear length and the crosses of UK 24, PL 28 and GRA with WPG 62528 showed good s.c.a. effects for grains per ear. The crosses which showed positive and significant s.c.a. effects for 250-grain weight were UK 27  $\times$  EC 70849, UK 24  $\times$  EB 4145, PL 28  $\times$  RD 47, PL 26  $\times$  WPG 62528, PL 26  $\times$  EC 70849 and GRA  $\times$  HBL 6. Out of these, PL 28  $\times$  RD 27 had significant effect for grain yield also. Another cross which exhibited significant effect for this trait was GRA  $\times$  EC 70849. The majority of the crosses that showed high s.c.a. effects involved at least one good general combiner. There were certain crosses in which both low  $\times$  low and high  $\times$  high combinations were operating. In the first type high genetic diversity and epistatic interaction among the parents led them to show high s.c.a. effects. The magnitude of s.c.a. effects in the second type was not to the extent of first type which might be due to some internal cancellation of favourable factors as suggested by JINKS and JONES (1958). The high s.c.a. effects of the crosses involving parents with high  $\times$  low general combining ability were due to dominant  $\times$  recessive interaction. Whenever good combiners gave low combining ability in combination, the involving parents were considered to be genetically similar. These findings lend support to the earlier observations of YAP and HARVEY (1971) in this crop.

The most successful method for exploiting sca effects is the heterosis breeding which is now a days technically possible in self-pollinated crops also (ATHWAL and BORLAUG 1967). Keeping this in view, the performance of the best four hybrids, inbreeding depression and their s.c.a. effects were studied (Table 5). It was interesting to note that the hybrid K 13  $\times$  HBL 6 which exhibited the highest heterosis of 60.84 per cent over the superior parent involved the parents with negative and low g.c.a. effects but had moderate s.c.a. effects in combination. The manifestation of heterosis in such cases may be regarded as arising due to epistatic interaction. On the other hand, the cross RD 47  $\times$  GRA involving parents with high g.c.a. effects for grain yield gave only 21.55 per cent increase. The s.c.a. effect was also negative which indicated that the involving parents may not be genetically diverse. The remaining two crosses viz., PL 28  $\times$  RD 47 and GRA  $\times$  HBL 6 with high sca effects comprised parents of high  $\times$  low general combining ability and gave 48.40 and 38.66 per cent increase for grain yield respectively. The hybrids in general, showed inbreeding depression for all the characters except 250-grain weight. The highest and lowest depression for grain yield was recorded for the crosses GRA  $\times$  HBL 6 and K 13  $\times$  HBL 6 respectively. These results indicated that K 13  $\times$  HBL 6 was the most promising combination as it also showed significant heterosis for other yield components like tillers per plant, leaf area, spikelets per spike and grains per ear and least inbreeding depression. However, it was tall, late flowering and late maturing type. The other two cross combinations PL 28  $\times$  RD 47 and GRA  $\times$  HBL 6 also need due consideration since they involved high  $\times$  low general combiners and registered substantial heterosis and s.c.a. effects.

Heterosis and combining ability for yield and its components were studied in a diallel involving ten inbreds of barley. Significant genotypic differences were observed among the parents and their hybrids for all the characters except 250-grain weight. The variances due to g.c.a. and s.c.a. were highly significant for all the characters except number of tillers per plant which was exclusively under the control of additive gene effect. High g.c.a.; s.c.a. ratio revealed

**Table 1**  
*Analysis of variance for different characters in a  $10 \times 10$  diallel cross of barley*

Source of variation	d.f.	Mean squares									
		Days to 70% heading	Days to maturity	Plant height	Tillers per plant	Leaf area	Spikelets per spike	Ear length	Grains per ear	250-grain weight	Grain yield
Replications	2	2.300	0.750	141.400	82.277	81.425	54.147	2.684	435.380	2.096	184.101
Progenies	54	118.333**	48.775**	286.890**	9.510**	281.863**	13.223**	3.122**	113.320**	4.440**	55.732**
Parents	9	245.855**	68.181**	288.276**	12.440**	287.267**	4.482**	8.339**	46.512**	7.242**	34.504**
Hybrids	44	992.965**	44.053**	184.102**	9.037**	282.442**	10.281**	2.055**	96.853**	3.315**	52.001**
Parents Vs. hybrids	1	87.100**	81.900**	4797.200**	3.963**	207.840**	221.346**	2.679	1439.150**	28.709**	410.970**
Error	108	2.325	0.819	17.268	4.663	24.046	1.514	0.712	18.656	0.375	16.732

\* Significant at  $P = 0.05\%$ \*\* Significant at  $P = 0.01\%$ 

**Table 2**  
*Analysis of variance for combining ability*

Source of variation	d.f.	Mean squares									
		Days to 70% heading	Days to maturity	Plant height	Tillers per plant	Leaf area	Spikelets per spike	Ear length	Grain per ear	250-grain weight	Grain yield
G.C.A.	9	212.592**	79.546**	169.583**	5.958**	268.186**	3.933**	4.085**	30.316**	5.659**	38.980**
S.C.A.	45	4.841**	3.596**	80.841**	2.612	59.109**	4.502**	0.431**	39.266**	0.644**	14.405**
Error	108	0.775	0.273	5.756	1.554	8.015	0.504	0.237	6.218	0.125	5.577
G.C.A. : S.C.A.		44.161 : 1	22.120 : 1	2.097 : 1	2.281 : 1	4.537 : 1	0.873 : 1	9.477 : 1	0.772 : 1	8.787 : 1	2.706 : 1

\* Significant at  $P = 0.05\%$ \*\* Significant at  $P = 0.01\%$

**Table 3**  
General combining ability effects of parents

Parents	Days to 70% heading	Days to maturity	Plant height	Tillers plant	Leaf area	Spikelets per spike	Ear length	Grain per ear	250-grain weight	Grain yield
UK 24	-5.616**	-2.725**	-6.398**	10.372**	-8.920**	-0.408*	0.146	-1.977**	0.294**	0.127
PL 28	-4.421**	-2.455**	2.001**	-0.199	-0.340	0.802**	1.262**	2.233**	2.531**	-0.580
PL 26	-2.727**	-2.372**	4.171**	-1.149**	3.440**	-0.272	0.232	-0.999	0.864**	1.258
RD 47	-2.477**	-1.566**	-1.803**	0.808*	-3.642**	-0.791**	-0.415**	-1.913**	0.744**	2.119**
K 13	-2.227**	-1.566**	2.168**	-0.221	1.826*	-0.416*	0.579*	-0.977	0.289**	-0.180
GRA	-0.813**	-0.705**	-2.442**	0.328	-2.309*	-0.552**	-0.031**	-0.630	0.164	2.641**
WPG 62528	2.742**	1.544**	3.646**	-0.480	0.287	0.224	-0.331*	0.722	-0.471**	-0.922
HBL 6	4.328**	2.572**	3.821**	-0.318	8.812	0.863**	-0.253*	2.503**	0.371**	0.580
EC 70849	5.439**	3.655**	-4.784**	0.117	-1.765**	0.205	-0.545**	0.736	-1.088**	-2.741**
EB 4145	5.772**	3.599**	-0.378	-0.257	2.609**	0.341	-0.642**	0.200	-0.957**	-2.663**
S.E. (gi) $\pm$ 0.241		0.143	0.657	0.341	0.775	0.194	0.133	0.682	0.096	0.646

\* Significant at  $P = 0.05\%$

\*\* Significant at  $P = 0.01\%$



**Table 4**  
*Specific combining ability effects*

Crosses	Days to 70% heading	Days to maturity	Plant height	Tillers per plant	Leaf area	Spikelets per spike	Ear length	Grains per ear	250-grain weight	Grain yield
UK 24 × PL 28	-7.062**	0.633	-5.868	-0.212	5.553	-0.560	-0.219	-1.101	-0.737*	-0.235
× PL 26	0.701	-0.449	-7.070	-0.662	-8.693	-0.183	-0.088	-1.001	-0.637	-0.940
× RD 47	-2.881	-1.255	-5.195	2.412*	6.456	0.469	-0.925*	1.645	-0.584	-2.335
× K 13	-1.465	-1.588	-1.534	1.176	-2.046	-1.471	-0.002	-3.223	0.537	2.603
× GRA	1.120	-0.449	-1.323	0.226	-11.210*	-0.769	-0.491	-3.104	0.562	1.676
× WPG 62528	0.434	-1.366	4.487	1.368	5.492	3.319*	0.841	10.809**	0.065	1.706
× HBL 6	-1.020	0.272	7.412	2.007	8.467	2.747	0.630	8.695	0.465	3.537
× EC 70849	-0.798	2.189	16.984**	0.304	0.711	2.272	0.489	7.395	0.915**	-1.907
× EB 4145	-0.798	2.224	4.779	-2.720**	-7.730	-0.260	0.253	-5.734	1.084**	-2.085
PL 28 × PL 26	1.840	-0.366	-5.170	-0.756	-10.870	-1.363	-1.738**	-4.179	-0.673	-2.865
× RD 47	-2.742	-3.550**	-0.462	1.551	-1.224	-0.144	-0.758	-0.865	1.112**	8.039**
× K 13	2.007	-2.171	1.765	-0.917	-10.293	-0.419	-0.185	-1.501	-0.232	-2.887
× GRA	-2.740	-2.366*	1.542	-0.001	2.042	0.016	0.191	-0.948	0.092	2.151
× WPG 62528	0.370	2.717*	5.354	0.907	3.844	3.105*	0.591	9.098*	0.362	-3.251
× HBL 6	-0.215	1.689	8.112	-1.820	10.653	0.333	-0.519	1.117	0.462	0.045
× EC 70849	-2.326	2.939	14.817**	-0.956	6.497	3.398**	0.672	8.717	0.379	-2.832
× EB 4145	-0.326	2.994	3.712	-1.881	4.556	1.789	-0.436	6.554	0.648	4.756
PL 26 × RD 47	-0.770	-0.921	-5.632	-0.031	-2.305	-0.966	0.405	-3.232	-0.220	-3.065
× K 13	2.979	0.078	2.962	2.465*	-5.507	0.491	0.244	1.331	-0.365	2.939
× GRA	-1.767	-0.116	6.006	-0.051	-0.138	1.228	-0.144	2.784	-0.107	-2.487
× WPG 62528	1.343	1.967	5.883	1.290	1.831	-0.583	-0.710	-1.701	1.729**	2.776
× HBL 6	-0.242	1.939	2.342	1.396	8.539	2.744	0.678	8.451	0.862	2.739
× EC 70849	-1.020	1.189	8.415	-1.106	8.483	1.936	0.069	6.084	0.979**	3.462
× EB 4145	-0.020	1.578	5.309	1.401	1.242	2.333	0.966**	7.954	-0.084	4.651

RD 47 × K 13	3.729**	3.606**	8.770	1.207	— 3.791	0.611	—0.808	0.912	0.544	4.412
× GRA	—0.351	—0.255	2.248	0.357	— 8.588	—0.452	—0.163	— 2.901	—0.220	—0.248
× WPG 62528	1.759	1.494	5.626	—2.001	6.314	1.703	0.879	4.545	0.248	—2.285
× HBL 6	1.173	1.133	3.551	—0.395	3.289	1.697	0.425	5.065	—0.184	—0.454
× EC 70849	0.062	1.383	9.423	—3.464**	7.667	0.455	0.383	2.531	0.331	3.267
× EB 4145	—0.604	1.106	13.584*	—1.489	3.492	1.653	0.414	2.601	0.634	0.256
K 13 × GRA	—0.267	—1.588	— 1.390	—2.312*	8.475	—0.427	0.575	— 2.637	—0.798*	—5.176*
× WPG 62528	—0.156	0.828	6.354	0.162	4.378	2.228	0.775	8.309	0.604	—0.346
× HBL 6	0.590	0.800	— 1.400	0.835	7.319	1.089	—0.069	0.495	0.404	3.551
× EC 70849	2.812	1.050	0.617	—0.967	10.231	0.347	—0.144	1.262	0.454	1.673
× EB 4145	1.479	1.772	9.079	0.940	— 0.371	1.744	0.486	5.631	0.756*	2.528
REA × WPG 62528	3.062*	1.300	3.131	0.079	1.514	3.397*	—0.046	9.129*	0.637	—0.207
× HBL 6	2.509	1.270	— 0.176	1.018	3.256	—0.774	—0.391	— 3.318	0.895**	3.223
× EC 70849	2.398	0.522	8.295	3.082**	1.067	0.883	—0.066	1.448	0.312	10.878**
× EB 4145	1.398	1.244	8.367	0.523	— 3.107	—0.512	—0.035	— 1.815	0.248	1.734
WPG 62528 × HBL 6	—0.045	—0.310	— 3.198	—0.073	—11.407	—1.552	—0.224	— 4.670	—0.001	2.253
× EC 70849	—0.823	—0.227	— 2.359	1.223	1.103	—3.027*	—0.366	— 9.304*	—0.084	0.976
× EB 4145	0.156	—0.671	— 0.832	—2.201*	6.561	—0.763	—0.302	— 1.968	—0.482	—1.801
HBL 6 × EC 70849	—1.076	—2.088	5.665	—0.670	— 6.855	—0.166	0.489	— 0.584	0.281	—0.960
× EB 4145	—0.409	—0.699*	3.926	—0.095	1.636	0.930	0.253	3.651	0.617	—0.437
EC 70849 × EB 4145	0.479	—2.116	—15.668**	2.235	—22.218**	—3.944**	—1.021*	—11.182**	—1.698**	—5.548*
S.E. (Sij) ±	2.02	1.74	8.28	1.47	7.08	1.95	0.61	5.77	0.46	3.49

\* Significant at P = 0.05%

\*\* Significant at P = 0.01%

Table 5

*Heterosis over better parent, inbreeding depression and sca effects of four best hybrids*

Crosses	Days to 70% heading			Days to maturity			Plant height			Tillers per plant			Leaf area		
	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.
PL 28 × RD 47	0.74	3.348	−2.742	−1.06	0.000	−3.550**	25.63**	6.109	−0.462	−4.65	9.357	1.551	0.92	6.050	−1.224
RD 47 × GRA	1.05	9.414	−0.351	1.58**	4.442	−0.255	23.32**	9.525	2.248	−9.59	9.417	0.357	19.04	13.985	−8.588
K 13 × HBL 6	14.60**	9.509	0.590	5.01**	5.505	0.800	14.81**	8.309	−1.420	28.50**	7.692	0.835	16.41**	9.156	7.319
GRA × HBL 6	10.87**	9.496	2.509	3.44**	4.477	1.270	13.01**	3.541	−0.176	22.72	11.212	1.018	5.67	1.007	3.256
Crosses	Spikelets per spike			Ear length			Grain per ear			250-grain weight			Grain yield		
	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.	Heterosis	Inbreeding depression	S.C.A.
PL 28 × RD 47	5.95	8.691	−0.144	−6.98	5.510	−0.758	4.91	3.528	−0.865	13.97**	2.836	1.112**	48.40**	7.142	8.039**
RD 47 × GRA	2.79	5.909	−0.452	−8.36	7.582	−0.163	−6.48	5.981	−2.901	0.26	−9.677	−0.220	21.55*	9.213	−0.248
K 13 × HBL 6	14.10**	4.015	1.089	−4.45	8.001	−0.069	14.30**	3.280	2.495	6.15	−5.000	0.404	60.84**	2.275	3.551
GRA × HBL 6	6.04	7.094	−0.774	−9.03	2.777	−0.391	−1.21	1.233	−3.318	4.48	−2.419	0.895**	38.66**	9.514	3.223

\* Significant at  $P = 0.05\%$ \*\* Significant at  $P = 0.01\%$



preponderance of additive genetic variance for all the characters except spikelets per spike and grains per spike where non-additive gene effects were more important. Inbreds RD 47 and GRA showed better combining ability for days to 70% heading, days to maturity, plant height and grain yield, while PL 28 and HBL 6 for spikelets per spike, grains per ear and 250-grain weight. The crosses K 13  $\times$  HBL 6, PL 28  $\times$  RD 47 and GRA  $\times$  HBL 6 have been identified as promising in terms of breeding value based on their heterosis and s.c.a. estimates as well as g.c.a. effects of the parents. The hybrids, in general, showed inbreeding depression in  $F_2$  generation.

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\*

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#### References

- ATHWAL, D. S. and BORLANG, N. E. (1967): Genetic male sterility in wheat breeding. *Indian J. Genet.*, **27**, 136–142.
- COMSTOCK, R. E., ROBINSON, H. F. and HARVEY, P. H. (1949): A breeding procedure designed to use both general and specific combining ability. *Agron. J.*, **41**, 360–367.
- GRIFFING, B. (1956b): Concept of general and specific combining ability in relation to diallel crossing systems. *Aust. J. Biol. Sci.*, **9**, 463–493.
- GULATI, S. C., TANDON, J. P., JAIN, K. B. L. and MURTY, B. N. (1969): Combining ability in a diallel cross of barley. *Indian J. Genet.*, **29**, 209–215.
- JINKS, J. L. and JONES, R. M. (1958): Estimation of components of heterosis. *Genetics*, **43**, 223–234.
- KATARKI, B. H. (1963): Studies on combining ability for yield and other quantitative characters in barley. M.Sc. thesis, I.A.R.I., New Delhi.
- SMITH, E. L. and LAMBERT, J. W. (1968): Evaluation of early generation testing in spring barley. *Crop. Sci.*, **4**, 90–93.
- SOLANKI, K. R. and BAKASHI, J. S. (1974): Combining ability in barley. *Indian J. Genet.*, **34**, 79–83.
- TANDON, J. P., JOSHI, A. B. and JAIN, K. B. L. (1968): Genetic analysis of yield in a six row and two row varietal cross in barley I. Genetics of yield and its primary components. *Indian J. Genet.*, **28**, 239–251.
- YAP, T. C. and HARVEY, B. L. (1971): Heterosis and combining ability of barley hybrids in densely and widely seeded conditions. *Can. J. Pl. Sci.*, **51**, 115–122.

#### GENE SYSTEMS GOVERNING YIELD AND ITS COMPONENT CHARACTERS IN RICE (*ORYZA SATIVA* L.)<sup>1</sup>

With the emergence of plant type concept, emphasis in rice agriculture has also shifted to building up high yielding varieties and their culture, replacing the conventional varieties. Apart from the well-known shortcomings in agrotechnology and socioeconomic constraints characterizing rice agriculture in India, lack of adequate genetic clarity on yield, and the various auxiliary characters to which it is attributed, has impeded the breeding of a suitable architectural pattern for attainment of higher production. For sustained rise in the genetic yield potential, recourse must be taken to production breeding based on sound principles of biometrical genetics.

<sup>1</sup> Part of Ph.D. thesis (Calcutta University, 1974) of M.K.S.

Yield by itself is a complex character and is governed by the action and interaction of a number of quantitative components. It is necessary to get a clear picture of the genetic architecture of yield and its components. The percentage of protein, of which augmentation in rice is also an urgent necessity (GOVINDSWAMY and GHOSH 1974), needs clarification in regard to the gene system. Little work has been done on this crop to study the genetic architecture of yield and its components except some I.R.R.I. reports (ANON. 1966, 1967) and those of SHIMURA (1967), WU (1968), LI and CHANG (1970) and SINHA (1974).

The present report includes analysis of diallel crosses pertaining to (i) a set of six parents, representing *indica* rice varieties as well as derived lines involving *indica* and *japonica* parents, and (ii) a set of seven parents, representing *indica* varieties and one dwarf high-yielding strain. The main object was to find the type of gene action governing the expression of yield, related characters and protein content in rice.

There were two diallel sets of crosses involving six and seven parental varieties. Combining ability analyses were carried out according to Method I, Model I of Griffing in the case of  $6 \times 6$  set (it includes one set of  $F_1$ 's and Reciprocal  $F_1$ 's) and according to Method 2, Model I of Griffing in the case of  $7 \times 7$  set, assuming that maternal effects were absent. The parents used for the two diallel sets are stated below:

*Parents in  $6 \times 6$  diallel set*

1. N<sub>22</sub>
2. SLO 16
3. ASD 1
4. ADT 27
5. LATISHAL
6. T<sub>141</sub>

*Parents in  $7 \times 7$  diallel set*

1. VIJAYA
2. T<sub>141</sub>
3. H<sub>4</sub>
4. LATISHAL
5. SLO 16
6. ASD 1
7. ADT 27

The trials were conducted in a randomized block design with three replications each. Each row comprised 12 plants. Ten plants per strain per block were subjected to the study. Recommended agronomic management practices were followed. Data were collected on heading date, plant height, number of total tillers, number of ear bearing tillers, panicle length, number of spikelets per panicle, number of grains per panicle, 100 grain weight, grain yield per plant, length : breadth ratio of grain and protein percentage.

**Table 1**

*Analysis of variance for the*

Source of variation	Diallel set	D.F.	Mean sum of squares			
			1	2	3	4
Between blocks	$6 \times 6$	2	8.55*	1.70	1.59	0.58
	$7 \times 7$	2	7.00*	23.30**	4.62*	3.97*
Between strains	$6 \times 6$	35	191.49**	211.24**	39.49**	39.08**
	$7 \times 7$	48	228.15**	324.89**	33.37**	41.40**
Error	$6 \times 6$	70	1.85	8.16	2.48	2.04
	$7 \times 7$	96	1.58	4.34	1.26	0.84

\* Significant at 5% level of probability; \*\* Significant at 1% level of probability.

1 = Heading date, 2 = Plant height, 3 = Total number of tillers, 4 = Number of ear bearing tillers, 5 = Panicle length, 6 = Number of spikelets/panicle, 7 = Number of grains/

Table 2

*Analyses of variance of combining abilities for different characters studied in rice*

Characters	Diallel set 6 × 6 7 × 7	g.c.a. 5 D.f. 6 D.f.	s.c.a. 15 D.f. 21 D.f.	Reciprocal 15 D.f.	Error 70 D.f. 96 D.f.	g.c.a. : s.c.a.
Heading date	6 × 6 7 × 7	385.24** 478.96**	19.68** 36.98**	0.83	1.85 1.58	19.57 : 1 12.95 : 1
Plant height	6 × 6 7 × 7	83.26** 160.98**	134.78** 201.52**	1.75	8.16 4.33	0.62 : 1 0.80 : 1
No. of total/tillers	6 × 6 7 × 7	17.93** 37.77**	23.64** 14.63**	1.09	2.48 1.26	0.76 : 1 2.58 : 1
No. of ear bearing tillers	6 × 6 7 × 7	16.75** 49.86**	24.08** 17.29**	0.74	2.04 0.84	0.70 : 1 2.88 : 1
Panicle length	6 × 6 7 × 7	3.44** 4.43**	2.94** 3.04**	0.09	0.30 0.25	1.17 : 1 1.46 : 1
Number of spikelets/panicle	6 × 6 7 × 7	3446.46** 394.13**	338.71** 389.81**	22.33	144.23 138.63	10.18 : 1 10.11 : 1
Number of grains/panicle	6 × 6 7 × 7	775.92** 1904.88**	394.31** 364.33**	48.83	105.59 37.32	1.97 : 1 5.23 : 1
100 grain weight	6 × 6 7 × 7	0.38** 0.64**	0.02** 0.03**	0.001	0.006 0.004	16.74 : 1 25.66 : 1
Yield/plant	6 × 6 7 × 7	45.79** 100.43**	44.07** 47.30**	3.82	3.87 2.50	1.04 : 1 2.12 : 1
Length/breadth ratio of grain	6 × 6 7 × 7	1.08** 1.28**	0.08** 0.05**	0.002	0.003 0.005	13.50 : 1 27.62 : 1
Protein percentage	6 × 6 7 × 7	6.63** 7.82**	0.72** 0.79**	0.01	0.03 0.002	9.19 : 1 9.93 : 1

\*\* = Significant at 1% level of probability

*different characters studied in rice*

Mean sum of squares						
5	6	7	8	9	10	11
0.03	59.00	33.30	0.002	9.50	0.003	0.373
2.39*	409.50	113.30*	0.001	17.01**	0.004	0.040**
5.37**	1941.30**	902.31**	0.19**	81.20**	0.57**	3.79**
5.66**	1989.60**	1192.53**	0.27**	99.74**	0.54**	3.97**
0.30	144.23	105.59	0.006	3.87	0.003	0.036
0.25	138.63	37.32	0.004	2.50	0.005	0.002

panicle, 8 = 100 grain weight, 9 = Yield/plant, 10 = Length/breadth ratio of grain and 11 = Protein percentage.



The analysis of variance (Table 1) revealed that mean sums of squares due to parents, hybrids and parents vs hybrids (referred to hereafter as lines) were significantly different. The mean sums of squares for both sets due to general and specific combining abilities are given in Table 2. Estimates of g.c.a. and s.c.a. effects are given in Tables 3 and 4 for  $6 \times 6$  and in Tables 5 and 6 for  $7 \times 7$  diallel sets, respectively. As the reciprocal effects were insignificant, their data have not been presented in this paper. Highly significant g.c.a. and s.c.a. mean sums of squares were recorded for all the characters (Table 7).

**Table 3**

*Estimates of g.c.a. effects of parents for different character in  $6 \times 6$  diallel cross of rice*

Char- acters	Parents							SE <sub>(gi)</sub>	SE <sub>(gi-gj)</sub>
	N 22	SLO 16	ASD 1	ADT 27	Latishal	T 141			
1	— 6.44**	—2.33**	—4.00**	—1.05**	6.30**	7.53**	0.36	0.56	
2	— 3.96**	1.65**	—1.95**	—0.44	1.55*	3.15**	0.75	1.17	
3	0.04	—0.26	2.15**	—1.02*	—1.27**	0.37	0.41	0.64	
4	0.0001	0.62	1.92**	—1.22**	—0.89*	0.81*	0.37	0.58	
5	— 0.93**	0.17	0.68**	—0.14	—0.03	0.25	0.14	0.22	
6	—23.20**	—5.25	—3.09	4.71	—2.03	28.88**	3.16	4.90	
7	— 1.15	0.12	—8.16**	2.65	—7.38**	13.93**	2.71	8.56	
8	0.07	—0.16**	0.22**	—0.22**	0.16**	—0.07**	0.02	0.03	
9	— 1.11*	—2.50**	2.03	—1.33*	0.61	2.30	0.52	0.80	
10	— 0.08**	0.41**	—0.12**	—0.47**	0.13**	0.14**	0.01	0.02	
11	— 0.32	0.84**	0.81**	1.01**	—0.28**	0.44**	0.05	0.06	

\* = Significant at 5% level of probability

\*\* = Significant at 1% level of probability

N.B. The notations 1–11 stand for different characters as mentioned in Table 1

**Table 5**

*Estimates of g.c.a. effects of parents for different characters in  $7 \times 7$  diallel cross of rice*

Char- acters	Parents								SE (gi)	SE (gi-gj)
	Vijaya	T 141	H 4	Latishal	SLO 16	ASD 1	ADT 27			
1	2.83**	3.87**	7.54**	3.48**	— 8.01**	—5.81**	—3.90**	0.31	0.47	
2	—6.81**	0.67	0.04	2.19**	2.66**	2.78**	—1.55**	0.51	0.78	
3	1.85	—0.15	— 1.49**	— 0.56*	— 0.65*	2.65**	—1.64**	0.27	0.42	
4	2.24	0.43	— 2.51**	— 0.40	— 0.58**	2.52**	—1.71**	0.22	0.34	
5	—0.21**	—0.34**	0.68**	— 0.49**	0.18	0.78**	—0.61**	0.12	0.19	
6	28.22**	18.84**	—15.55**	—10.69**	—11.86**	—4.06	—4.87	2.91	4.45	
7	21.30**	5.05**	—12.75**	—10.65**	5.23**	—4.02**	—4.15**	1.51	2.30	
8	0.08**	—0.15**	0.27**	0.07**	— 0.16**	0.20**	—0.31**	0.01	0.02	
9	4.76**	0.08**	— 2.37**	— 0.45	— 0.24	1.64**	—3.42**	0.39	0.59	
10	0.11**	0.08**	0.03*	0.06**	0.45**	—0.20**	—0.53**	0.02	0.03	
11	—0.06**	—0.54**	— 0.16**	— 0.30**	0.67**	—0.90**	1.29	0.01	0.02	

\* gi = Significant at 5% level of probability

\*\* gi = Significant at 1% level of probability

N.B. The notations 1–11 stand for different characters as mentioned in Table 1

Table 4

*Estimates of s.c.a. effects of rice crosses for different characters in 6×6 diallel set*

Crosses	1	2	3	4	5	6	7	8	9	10	11
N 22×SLO 16	-0.86**	-7.20**	2.41**	1.77**	1.32**	-10.49**	-6.45**	0.03**	2.06**	-0.17**	-0.08**
N 22×ASD 1	2.14**	6.09**	2.49**	2.25**	0.68**	4.18**	-9.01**	0.04**	0.05**	0.07**	0.14**
N 22×ADT 27	0.36**	1.46**	3.49**	3.83**	0.28**	-5.80**	8.35**	0.06**	4.26**	-0.01**	-0.64**
N 22×Latishal	-1.00**	2.28**	-1.09**	-1.27**	-0.54**	6.79**	10.71**	0.10**	-0.01**	-0.08**	0.22**
N 22×T 141	-1.39**	8.34**	1.43**	1.36**	-0.46**	14.70**	25.24**	0.02**	3.87**	0.09**	21.24**
SLO 16×ASD 1	4.19**	2.95**	1.29**	1.52**	0.16*	13.40**	0.21	0.04**	1.92**	-0.16**	-0.61**
SLO 16×ADT 27	3.92**	11.52**	1.13**	1.50**	1.03**	23.26**	10.07**	0.08**	1.62**	-0.33**	1.37**
SLO 16×Latishal	4.27**	5.44**	1.88**	1.66**	0.71**	7.51**	8.94**	0.01	1.43**	0.04**	-0.49**
SLO 16×T 141	-3.83**	8.71**	0.74**	0.97**	1.51**	6.92**	11.96**	0.05**	0.70**	-0.17**	-0.51
ASD 1×ADT 27	-2.74**	-5.27**	-0.45*	-0.86**	-0.22**	-8.07**	12.85**	0.04**	-0.21	0.01	-0.55**
ASD 1×Latishal	-4.28**	4.93**	2.46**	2.80**	0.45**	0.18	2.54*	0.04**	4.99**	-0.05	0.20**
ASD 1×T 141	-0.83**	-2.39**	2.49**	2.77**	-0.02	-3.40*	-9.09*	-0.12**	1.98**	0.16**	0.32**
ADT 27×Latishal	2.11**	5.15**	0.46**	0.61**	-0.23**	-0.29	-11.43**	-0.22**	-3.19**	0.08**	-0.48**
ADT 27×T 141	-1.61**	-2.94**	-0.68**	-0.92**	0.39**	4.45**	-4.06**	-0.04**	1.68**	-0.01	-0.71**
Latishal×T 141	1.86***	-3.67**	0.74**	0.75**	-0.22**	-5.46**	2.29**	-0.05**	3.88**	0.02**	0.30**
SE (Sij)	0.16	0.33	0.18	0.16	0.06	1.41	1.21	0.01	0.23	0.01	0.02
SE (Sij-Sik)	1.24	2.60	1.43	1.30	0.50	10.96	9.38	0.01	1.79	0.05	0.17
SE (Sij-Skl)	1.11	2.33	1.29	1.16	0.44	9.80	8.39	0.01	1.60	0.04	0.15

\* Sij = Significant at 5% level of probability

\*\* Sij = Significant at 1% level of probability

N.B. The notations 1-11 stand for different characters as mentioned in Table 1

Table 6

*Estimates of s.e.a. effects of rice crosses for different characters in 7 × 7 diallel set*

Crosses	Characters										
	1	2	3	4	5	6	7	8	9	10	11
A × B	-2.54**	3.52*	-0.90**	-0.95**	0.01	17.67**	-1.82**	-0.03**	0.57**	-0.04**	0.28**
A × C	-1.44	0.42	-1.76**	-1.67**	-0.58**	-2.75**	-12.31**	-0.12**	-1.32**	-0.01	-0.13**
A × D	1.24	3.96*	0.92**	0.46**	-0.41**	-9.71**	-22.45**	-0.12**	-4.31**	-0.03**	0.19**
A × E	-2.29**	0.27	0.40**	0.36**	1.34**	11.32**	20.19**	-0.01	6.01**	0.14**	-0.23**
A × F	3.67**	16.20**	1.09**	1.75**	1.02**	6.62**	1.45**	0.03**	1.36**	-0.02*	-0.05**
A × G	3.50**	12.83**	4.43**	4.39**	0.35**	-5.04**	-11.18**	0.02*	9.75**	0.01	-0.41**
B × C	2.21*	-3.54**	-0.06	0.43**	0.48**	5.05**	-1.93	-0.07**	-1.31**	0.08**	-0.13**
B × D	0.50	-1.60	0.33**	0.03	-0.29**	-8.67**	-0.36	-0.02**	4.44**	0.02*	0.47**
B × E	-2.26*	8.92	1.48**	1.24**	1.20**	3.86**	13.57**	-0.02*	0.73**	-0.30**	-0.42**
B × F	-0.29	0.03	1.88**	2.50**	-0.57**	8.60**	-0.12	-0.09**	1.20**	0.16**	0.37**
B × G	-1.63	2.25	1.77**	1.23**	0.03	-3.52**	-2.99**	0.02**	1.48**	0.04**	-1.07**
C × D	0.30	3.08	0.06	0.55**	0.49**	15.39**	16.10**	0.05**	0.60**	-0.03*	-0.26**
C × E	-6.00**	0.98	1.34**	1.02**	0.79**	4.90**	5.74	0.14**	3.18**	-0.01	-0.63**
C × F	3.60**	11.12**	3.51**	5.08**	1.08	18.66**	4.54**	0.15**	5.21**	-0.01	0.01
C × G	4.32**	-2.72	-3.24**	-4.45**	-0.23**	17.72**	-5.72**	-0.09**	-6.34**	0.02*	1.50**
D × E	5.68**	3.63	1.36**	1.06**	0.88**	-1.02	-2.81**	0.08**	3.54**	0.02*	-0.32**
D × F	4.30**	0.03	2.39**	1.18**	0.70**	-0.89	-1.98**	-0.01	2.34**	0.02*	0.21**
D × G	1.04	3.61**	-0.38**	0.75**	-0.39**	1.51	-5.49**	-0.16**	-2.19**	0.01	-0.56**
E × F	6.71**	-1.02	0.27*	0.15	-0.45**	-14.52**	-17.61	0.01	-1.23**	-0.20**	-0.15**
E × G	5.64**	9.61**	1.31**	2.12**	1.55**	26.95**	1.88	0.06**	-0.45**	-0.27**	1.15**
F × G	-6.55**	-11.47**	-1.36**	-1.85**	-0.32**	1.44	22.07*	0.02**	-0.46**	0.02*	-0.92**
SE (Sij)	1.07	1.78	0.11	0.09	0.05	1.18	0.61	0.01	0.16	0.01	0.01
SE (Sij-Sik)	1.16	1.92	1.03	0.84	0.46	10.90	5.66	0.05	1.46	0.07	0.04
SE (Sij-Skl)	1.06	1.75	0.94	0.77	0.42	9.95	5.16	0.05	1.33	0.05	0.03

\* Sij = Significant at 5% level of probability

\*\* Sij = Significant at 1% level of probability

A = Vijaya, B = T 141, C = H 4, D = Latishal, E = SLO 16, F = ASD 1 and G = ADT 27

N.B. The notations 1-11 stand for different characters as mentioned in Table 1



Table 7

Best parents, best general combiners and best specific combiners resulting from analysis of two diallel sets in rice

Characters	Diallel set	Best parent	Best F <sub>1</sub>	Best s.c.a.	Best g.c.a.
Heading date	6×6	T 141	T 141×Latishal	SLO 16×ASD 1 = +Highest	T 141 = +Highest
				SLO 16×Latishal = -Highest	N 22 = -Highest
	7×7	(H 4)	(Latishal×H 4)	SLO 16×ASD 1 = +Highest	H 4 = +Highest
				SLO 16×Latishal = -Highest	SLO 16 = -Highest
Plant height	6×6	T 141	SLO 16×T 141	SLO 16×ADT 27 = +Highest	T 141 = +Highest
				SLO 16×N 22 = -Highest	N 22 = -Highest
	7×7	(T 141)	(H 4×ASD 1)	Vijaya ×ASD 1 = +Highest	ASD 1 = +Highest
				ASD 1 ×ADT 27 = -Highest	Vijaya = -Highest
Total tillers	6×6	ASD 1, T 141	T 141×ASD 1	N 22×ADT 27	ASD 1
	7×7	(Vijaya)	(Vijaya×ASD 1)	(Vijaya×ADT 27)	(ASD 1)
Ear-bearing tillers	6×6	T 141	T 141×ASD 1	N 22×ADT 27	ASD 1
	7×7	(Vijaya)	(Vijaya×ASD 1)	(H 4×ASD 1)	(ASD 1)
Panicle length	6×6	ASD 1	T 141×SLO 16	SLO 16×T 141	ASD 1
	7×7	(ASD 1)	(H 4×ASD 1)	(SLO 16×ADT 27)	(H 4)
No. of spikelets/panicle	6×6	T 141	T 141×ADT 27	SLO 16×ADT 27	T 141
	7×7	(Vijaya)	(Vijaya×T 141)	(SLO 16×ADT 27)	(Vijaya)
No. of grains/panicle	6×6	T 141	N 22×T 141	N 22×T 141	T 141
	7×7	(Vijaya)	(Vijaya×T 141)	(ASD 1×ADT 27)	(Vijaya)
100 grain weight	6×6	Latishal	ASD 1×Latishal	N 22×Latishal	ASD 1
	7×7	(H 4)	(H 4×ASD 1)	(H 4×ASD 1)	(H 4)
Yield/plant	6×6	ASD 1	ASD 1×Latishal	ASD 1×Latishal	T 141
	7×7	(Vijaya)	(Vijaya×ADT 27)	(Vijaya×ADT 27)	(Vijaya)
Length/breadth ratio	6×6	SLO 16	SLO 16×Latishal	ASD 1×T 141	SLO 16
	7×7	(SLO 16)	(Vijaya×SLO 16)	(T 141×ASD 1)	(SLO 16)
Protein percentage	6×6	ADT 27	SLO 16×ADT 27	SLO 16×ADT 27	ADT 27
	7×7	(ADT 27)	(SLO 16×ADT 27)	(SLO 16×ADT 27)	(ADT 27)

### *Days to heading*

The g.c.a. mean sums of squares were 19.57 and 12.90 times greater than s.c.a. mean sums of squares in  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively. This indicated greater role of additive genetic system in the inheritance of this character.

The highest positive and negative g.c.a. effects were observed in  $T_{141}$  and  $N_{22}$  in  $6 \times 6$  diallel set and in  $H_4$  and SLO 16 in  $7 \times 7$  diallel set. The cross SLO 16  $\times$  ASD 1 showed highest significant s.c.a. effect in both the diallel sets.

### *Plant height*

The g.c.a. mean sums of squares were 0.62 and 0.80 times less than s.c.a. mean sums of squares in  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively, suggesting greater prevalence of non-additive effects than additive effects of gene systems.

The g.c.a. effects of all the parents were significant, except ADT 27 in  $6 \times 6$  diallel set and  $T_{141}$  and  $H_4$  in  $7 \times 7$  diallel set. The highest positive and negative significant g.c.a. effects, respectively, were observed in  $T_{141}$  and  $N_{22}$  ( $6 \times 6$ ), and ASD 1 and Vijaya ( $7 \times 7$ ). The crosses SLO 16  $\times$  ADT 27 ( $6 \times 6$ ) and Vijaya  $\times$  ASD 1 ( $7 \times 7$ ) showed highest positive significant s.c.a. effects.

### *Total tillers*

The g.c.a. mean sums of squares were 0.76 times less ( $6 \times 6$ ) and 2.58 times greater ( $7 \times 7$ ) than s.c.a. mean sums of squares, suggesting that low non-additive effect had slightly greater role than additive effect in the case of  $6 \times 6$  diallel set and *vice versa* for  $7 \times 7$  diallel set, in respect of this character.

The highest s.c.a. value was observed in ASD 1 in both the diallel sets. The s.c.a. effects were significant, except in two crosses of  $7 \times 7$  diallel set viz.  $T_{141} \times H_4$  and  $H_4 \times$  Latishal. The highest positive s.c.a. effects were exhibited by  $N_{22} \times$  ADT 27 ( $6 \times 6$ ) and Vijaya  $\times$  ADT 27 ( $7 \times 7$ ).

### *Ear bearing tillers*

The g.c.a. mean sums of squares were 0.07 times less ( $6 \times 6$ ) and 2.88 times greater than s.c.a. mean sums of squares in  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively, proving that the non-additive effect had a greater role than the additive effect in the case of  $6 \times 6$  diallel set, and *vice versa* for  $7 \times 7$  diallel set.

Only the parents  $T_{141}$  and ASD 1 ( $6 \times 6$ ), and Vijaya and ASD 1 ( $7 \times 7$ ) had significantly positive g.c.a. effects. ASD 1 registered itself as the best general combiner in both the sets.  $N_{22} \times$  ADT 27 ( $6 \times 6$ ) and  $H_4 \times$  ASD 1 ( $7 \times 7$ ) had the highest s.c.a. scores in positive direction.

### *Panicle length*

The higher variances of g.c.a. over s.c.a. showed the predominance of additive effects. In  $6 \times 6$  set, only ASD 1 had a positive significant effect. In  $7 \times 7$  set, the highest positive significant g.c.a. effect was recorded in ASD 1 followed by  $H_4$ . The highest s.c.a. effects were recorded in SLO 16  $\times$   $T_{141}$  ( $6 \times 6$ ) and SLO 16  $\times$  ADT 27 ( $7 \times 7$ ).

*Spikelets/panicle*

The g.c.a. mean sums of squares were 10 times greater than s.c.a. mean sums of squares in both sets, suggesting significant additive effects. The highest significant positive g.c.a. effects were recorded in  $T_{141}$  ( $6 \times 6$ ) and Vijaya ( $7 \times 7$ ). The highest positive s.c.a. effects were given by SLO  $16 \times$  ADT 27 in both the sets.

*Grains/panicle*

The g.c.a. mean sums of squares were 1.97 and 5.23 times greater than s.c.a. mean sums of squares in  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively, signifying the prevalence of additive effect. The highest positive significant g.c.a. effects were marked in  $T_{141}$  and Vijaya for  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively.  $N_{22} \times T_{141}$  and ASD  $1 \times$  ADT 27 were the best specific combiners for  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively.

*100 grain weight*

The greater intensity of g.c.a. mean sums of squares than s.c.a. mean sums of squares in both sets indicated the prevalence of additive effect. The rank orders of good g.c.a. effects were ASD 1, Latishal,  $N_{22}$ ,  $T_{141}$ , SLO 16 and ADT 27 in  $6 \times 6$  set; and  $H_4$ , ASD 1, Vijaya, Latishal  $T_{141}$ , SLO 16 and ADT 27 in  $7 \times 7$  diallel set. The crosses showing significantly negative s.c.a. effects for  $6 \times 6$  diallel set were ASD  $1 \times T_{141}$ , ADT 27  $\times$  Latishal, ADT 27  $\times T_{141}$  and Latishal  $\times T_{141}$ ; otherwise all other crosses had positive g.c.a. effects. These proved that the crosses between the parents with higher grain weight had a general tendency to show lower s.c.a. effects. The best specific combining abilities were recorded for  $N_{22} \times$  Latishal and  $H_4 \times$  ASD 1 for  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively. This indicates that crosses between high and low grain weight parents were the best specific combiners.

*Yield/plant*

The g.c.a. mean sums of squares were 1.04 and 2.12 times greater than s.c.a. mean sums of squares. This suggested that the additive genetic component was playing a greater role in the inheritance of yield/plant. The rank orders were  $T_{141}$ , ASD 1, Latishal,  $N_{22}$ , ADT 27 and SLO 16 for  $6 \times 6$  diallel set; and ASD 1,  $T_{141}$ , SLO 16, Latishal,  $H_4$  and ADT 27 for  $7 \times 7$  diallel set for higher g.c.a. effects. The highest values for s.c.a. effects were recorded in ASD  $1 \times$  Latishal ( $6 \times 6$ ) and Vijaya  $\times$  ADT 27 ( $7 \times 7$ ).  $N_{22} \times$  ASD 1,  $N_{22} \times T_{141}$  and  $N_{22} \times$  SLO 16 ( $6 \times 6$ ); and Vijaya  $\times$  SLO 16,  $T_{141} \times$  Latishal,  $H_4 \times$  ASD 1 and Latishal  $\times$  SLO 16 ( $7 \times 7$ ), were recorded as the good specific combiners.

*Length/breadth ratio of grain*

Considerably greater g.c.a. mean sums of squares than s.c.a. mean sums of squares suggested the additive nature of gene action in both sets. The highest g.c.a. effect was marked in SLO 16 for both the sets. The highest s.c.a. effect was also observed for the ASD  $1 \times T_{141}$  cross combinations.

*Percentage of protein*

The g.c.a. mean sums of squares were 9.19 and 9.93 times greater than s.c.a. mean sums of squares in  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively, indicating the additive nature of



gene action. The high positive g.c.a. effects in order of merit in both sets were shown by ADT 27 and SLO 16; otherwise all other parents had negative g.c.a. effects. Few good specific combiners in order of merit were SLO 16  $\times$  ADT 27, ASD 1  $\times$  T<sub>141</sub> (6  $\times$  6); and H<sub>4</sub>  $\times$  ADT 27, and SLO 16  $\times$  ADT 27 (7  $\times$  7).

The present investigations showed that the total genetic variation for all the characters was associated with both additive and non-additive genetic effects. However, the magnitude of additive components was higher in almost all the cases except in plant height of both sets, and total and ear-bearing tillers of 6  $\times$  6 set. Thus, the variation in the expression of combining ability between the two diallel sets of the same species may probably be due to presence or absence of interacting parents in a particular set. CRUMPACKER and ALLARD (1962), too, have reported on such interaction effects in diallel crosses in wheat. All the strains of 6  $\times$  6 diallel set were long statured and in 7  $\times$  7 set one strain Vijaya had a dwarf plant type. The inclusion of Vijaya in the latter set might have changed the nature of the combining ability effect. Besides these, ear-bearing tillers, which act as direct auxiliary vehicles (SINHA 1974), were under the control of genes having a preponderantly additive effect in 7  $\times$  7 diallel set. It shows that this character may be manipulated in a better way to increase yield potential in the 7  $\times$  7 set. Thus, it may be broadly concluded that the utilization of dwarf plant types which improved the yield potential more than that obtainable in *indica* rice, might have facilitated accumulation of additive genes in governing expression of ear-bearing tillers and number of grains/panicle. In the event of the latter character, g.c.a. : s.c.a. ratio was higher in 7  $\times$  7 than in 6  $\times$  6 set.

As the magnitude of additive genetic components was higher for all the desirable characters, the selection work in these crosses can be justifiably handled through routine pedigree method. The plant height was governed by non-additive effect. This naturally calls for elimination of tall segregants in early generations. However, the results indicate that for achieving maximum grain production in rice both additive and non-additive genetic components have to be suitably manipulated. Taking into consideration the existence of different types of gene action in the population, the most effective breeding programme in rice improvement work appears to be one which, besides utilizing fixable gene effects, would also maintain considerable homozygosity for exploiting the dominance gene effects. It thus appears reasonable to take recourse to recurrent selection and biparental matings, as well as matings of selected plants in early segregating generations with a view to building a rice population having optimum homozygous and heterozygous balance. The procedure suggested by FREY (1975) as an alternative to conventional methods appears to hold the key to improvement under such situations. The requirement for successful implementation of this idea are (i) developing populations from a breed base of elite genetic stock, (ii) effective massive gene flow through extensively random or systematically diverse recombination, (iii) selecting for desired simply-inherited characters, (iv) evaluating performance of F<sub>1</sub>'s to F<sub>2</sub>'s for complex quantitatively-inherited characters under a range of environments, and (v) providing a continuing offtake of elite genotypes into "Pedigree" advancement and infusing new genetic materials into the system. However, the major limiting factors in adoption of population improvement technique in self-fertilizing crops like rice are these two: One relates to the question of ensuring sufficient quantity of seeds for multilocation testing. To overcome these difficulties RACHIE and GARDNER (1975) suggested increasing the rapidity and efficiency of hand crossing; introduction of some form of out-crossing like genetic male sterility, delayed dehiscence or protogyny into the population; and reducing seed number and plot size required for testing. The difficulty of hand crossing may be overcome by introducing the vacuum emasculator developed at International Rice Research Institute, Los Banos, Philippines.

The best general combiners in regard to yield were T<sub>141</sub> and ASD 1 in 6  $\times$  6 diallel set.

Both also ranked as best general combiners for five and four yield component characters, respectively. In case of  $7 \times 7$  diallel set, Vijaya, ASD 1 and  $T_{141}$  proved their mark as best general combiners not only for yield but also generally for those components that have registered maximum direct contribution to yield, viz. ear-bearing tillers and number of grains per panicle (SINHA 1974). High general combining ability effects are related to additive genetic effects or additive  $\times$  additive interaction effects (GRIFFING 1956) which represent the fixable genetic component of variation. Thus, crosses involving these or similar varieties are expected to produce segregates with near optimum levels of components of yield. It may be noted here that ASD 1 and  $H_4$  had the highest g.c.a. for grain weight in  $6 \times 6$  and  $7 \times 7$  diallel sets, respectively, but the reverse was true for number of grains/panicle. This may be due to negative association between the two characters (SINHA 1974). SLO 16 and ADT 27 had the highest g.c.a. value for protein percentage; besides these, SLO 16 was also the best combiner for length/breadth ratio of the grain. Hence, SLO 16 comes an important variety for inclusion in breeding programmes aimed at quality rice.

In contrast to general combining ability effects, specific combining ability effects represent dominance and epistatic components of variation which are non fixable and hence would not contribute tangibly in the improvement of self-pollinated crops, except in cases where commercial exploitation of heterosis is feasible. But the crosses showing high s.c.a. values, whose parents are good general combiners, could be exploited in rice breeding. In the present study, Vijaya  $\times$   $T_{141}$ , Vijaya  $\times$  ASD 1 ( $7 \times 7$ ) were such cross combination, despite the fact that these combinations did not register as the best specific combiners. In some cases, the specific combining abilities were better due to one parent being a good general combiner for yield and the other parent being a good general combiner for a direct contributor to yield. Such crosses were Vijaya  $\times$  SLO 16, and  $H_4 \times$  ASD 1 ( $7 \times 7$ ). In some cases where the specific combining ability is high but involves one good combiner, the other being less so, such a combination may throw up desirable transgressive segregates, if the additive genetic system present in the good-combiner and complementary epistatic effect in the lesser-combiner parent acted in the same direction to maximize the expression of the desirable attribute. In this study, the crosses ASD 1  $\times$  Latishal,  $N_{22} \times$  ADT 27,  $T_{141} \times$  Latishal and  $N_{22} \times T_{141}$  ( $6 \times 6$ ), and Vijaya  $\times$  ADT 27 and  $H_4 \times$  SLO 16 ( $7 \times 7$ ) proved to be very high specific combiners for grain yield. It is interesting to note that Vijaya  $\times$  ADT 27 ( $7 \times 7$  set) had the highest specific combining ability among all crosses. This cross combination possessed good general combiners for yield and protein. This offers a good promise of breeding for high yield, which simultaneously increases the nutritional status of consumable rice.

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## References

- CRUMPACKER, D. W. and ALLARD, R. W. (1962): A diallel cross analysis of heading date in wheat. *Hilgardia*, **32**, 274–318.
- FREY, K. J. (1975): Breeding concepts and techniques for self-pollinated crops. Proc. Int. Symp. Grain Legume, ICRI SAT, Hyderabad, India, 257–278.
- GOVINDSWAMY, S. and GHOSH, A. K. (1974): Breeding for high protein content in rice. *Indian J. Genet.*, **34**, 628–641.



- GRIFFING, B. (1956): Concept of general and specific combining ability in relation to diallel crossing. *Aust. J. Biol. Sci.*, **4**, 463–493.
- ANONYMOUS (1966): Annual Report. International Rice Research Institute, Los Banos, Phillipines.
- DO, H. (1967): Annual Report. International Rice Research Institute, Los Banos, Phillipines.
- LI, C. C. and CHANG, T. T. (1970): Diallel analysis of agronomic traits of rice. *Bot. Bull. Acad. Sinica*, **11**, 61–78.
- RACHIE, K. O. and GARDNER, C. O. (1975): Increasing efficiency in breeding partially out-crossing grain legumes. *Proc. Int. Symp. Grain Legumes*, ICRISAT, Hyderabad, India, 285–297.
- SINHA, M. K. (1974): Genetical studies in *Oryza* species. Ph.D. Thesis, Calcutta University (Unpublished).
- SHIMURA, E. (1967): Diallel analysis of varietal differentiation in a rice variety. *Jap. J. Breed.*, **17**, 157–164.
- WU, H. P. (1968): Studies on quantitative inheritance of *Oryza sativa* L. II. A diallel analysis for panicle number, tiller number, panicle length, spikelet number and number of primary branches in  $F_1$  progeny. *Bot. Bull. Acad. Sinica*, **9**, 124–138.

#### LIGHT INTERCEPTION, CANOPY TEMPERATURE AND PHOTOSYNTHESIS IN A YELLOW-GREEN MUTANT OF DURUM WHEAT

Morphological characteristics influence the manner in which canopies interact with the environment. Canopy structure influences the light, temperature and humidity conditions in which leaves develop and may therefore affect factors as photosynthetic and transpirational rate of leaves.

Differences in canopy structures influence the absorption and reflection of radiation by the plants.

Canopy temperature consequently gives an indication of canopy-environmental interactions. The differences of temperature among different plant canopies might result from a difference in net radiation absorption, sensible and latent heat flux. In cooler canopies latent heat flux should be diminished and therefore lower transpirational loss should be observed.

Leaf size, degree of waxiness, awn and plant color are morphological characteristics that influence radiation absorption and sensible heat flux [8]. GATES *et al.* [9] demonstrated that leaf color influences light reflection from plant surface.

In visible range, more radiation is reflected by light colored plants than by green plants. Moreover waxy plants reflected more radiation than non-waxy plants [3].

Increase in albedo should reduce plant surface temperature and evaporation [14].

Light-colored and awned barley lines demonstrated lower canopy temperatures than their green or awnless isogenic pairs [8]. BENCI *et al.* found in awned plants of barley a higher sensible heat exchange than in awnless plants [4].

SINGH [15] has shown that the low efficiency in dry matter production of light-green harley was compensated by the increased leaf surface. Therefore no significant difference was found in plant and spike dry weight of light-green lines compared with normal green cultivars.

In different species, light-green mutants usually show total chlorophyll content a  $1/4 : 1/2$  with respect to green control [1, 10, 12, 13] and chlorophyll a/b ratio is frequently higher than in green plants [2, 10, 1]. Photosynthetic rates of these mutants expressed on area basis are equivalent to their green control, however, much greater photosynthetic rates are found on chlorophyll basis [5, 6, 7, 10].

In this work the influence of pigment deficiency of a *durum* wheat mutant on solar radiation interception was studied through the canopy and air temperature inside the canopy.



Moreover, experiments were carried out for measuring photosynthetic and transpirational rates of flag leaf. No difference was detected between yellow-green mutant and green control on grain yield (about 40 q/ha).

Yellow-green mutant and green control (cv. Cappelli) were grown in one field: the plot size was 34 m<sup>2</sup>, seed investment 200 kg/ha<sup>-1</sup>, nitrogen supply 120 kg N/ha<sup>-1</sup>.

In order to measure transmission of total radiation through yellow-green and wild type canopies, groups of solarimeters Lintronic (400–3500 nm) were inserted at three sites in each of the varieties at 10 and 45 cm above ground level.

Reflections of total and infrared radiations were measured at three sites in each of the varieties by mounting, 2 m above ground level, groups of three solarimeters Lintronic. Separation of the visible and infrared radiation was obtained by using a Kodak Wratten filter 88A.

Measurements of temperature were made with nickel resistance thermometers (30 × 0.5 cm; 1005 at 0 °C) inserted at three sites in each of the varieties at 10 and 45 cm above ground level. The percentage transmissions at different wavelengths were determined with a ISCO spectroradiometer Mod. SR. All the information on the experiments was collected on seven days between 19 May and 6 June.

CO<sub>2</sub> exchange rate was determined on detached leaves by an open system [11] connected with a Beckman differential CO<sub>2</sub> analyzer Mod. 865. Transpirational rate was measured by a psychrometer Shaw, and chlorophyll a and b by McKinney 1 method in 80% acetone, using the specific absorption coefficients of McKinney compiled by ARNON. During measurements and analysis the average height of the yellow-green plants as well as that of the green control are similar (about 150 cm).

Mean values of percentage transmission through the canopies of total solar energy are shown in Table 1.

Table 1  
*Percentage transmission\**

Height** cm	Green control	YG
10	14.0	18.7
45	19.7	24.3

\* Mean irradiance during the measurements period was 44.5 mW/cm<sup>2</sup>

\*\* Height above ground level

At both heights, YG shows higher transmission values than the green control. In Fig. 1 radiation interception profiles are represented (at 10 cm above ground level) for both green and YG canopies.

Spectral analysis of radiation transmission through a single leaf confirms the above data (Fig. 2). The more relevant difference was found in PAR range. No difference was detected on near-infrared range (700–1500 nm).

#### *Abbreviations:*

YG = yellow-green  
PAR = Photosynthetic active radiation  
IR = Infrared

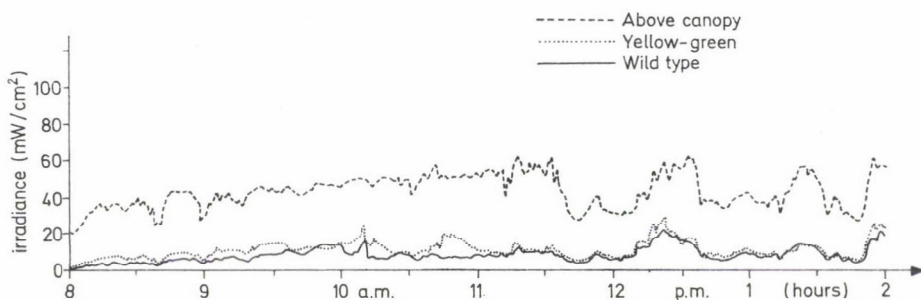


Fig. 1. Radiation transmission through green and yellow-green canopies on 24 May. Measurements inside the canopies were carried out by means solarimeters inserted at 10 cm above ground level

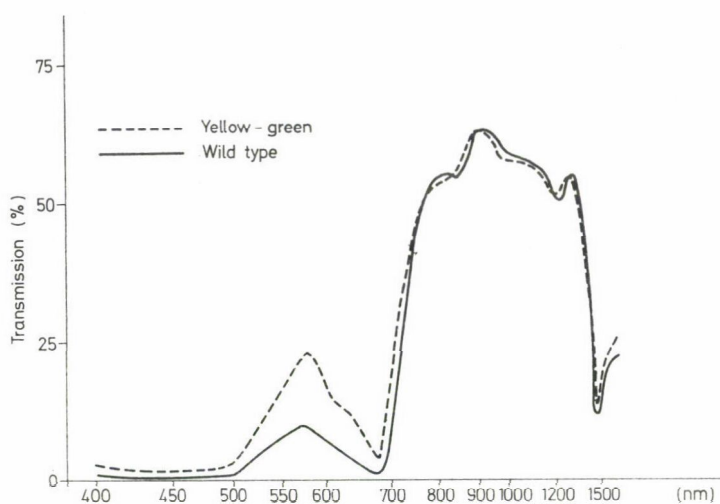


Fig. 2. Percentage transmission of PAR and near infrared solar radiation through a single leaf layer

Measurements of reflected radiation were made on both PAR and IR ranges. Experimental values are shown in Table 2. Higher reflection was observed on the YG canopy than on the green, when both PAR and IR radiations were measured.

Table 2

Percentage reflection

	Green control	YG
PAR	4.3	8.1
IR	44.3	50.6

Experimental data shown in Table 1 should indicate that the net absorption of radiation through the YG canopy is lower with respect to the green.

This phenomenon is correlated with light color of yellow-green mutants [7].

Because of differences of energy absorption by the yellow-green canopy with respect to the green, lower air temperatures inside the canopy were found (Table 3) at two different distance from ground level.

**Table 3**  
*Temperature ( $^{\circ}\text{C}$ )  
inside the canopy*

Height cm	Green control	YG
10	28.1	26.0
45	31.3	30.5

Experimental data represent mean values of temperature between 7 a.m. and 7 p.m. on seven days.

The diurnal pattern of temperatures at 10 cm from the ground level, determined during one day is shown in Fig. 3.

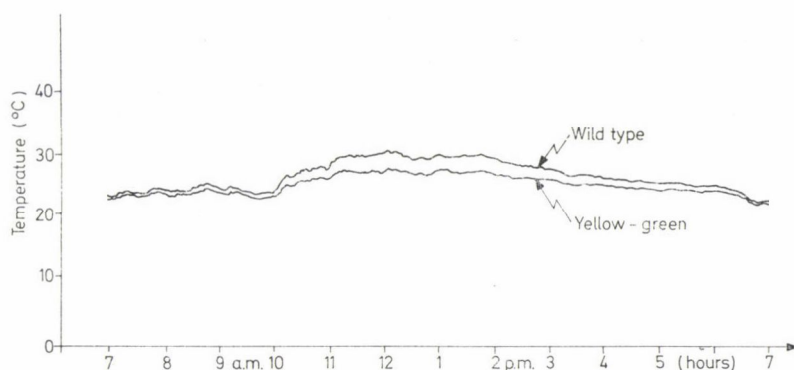


Fig. 3. Air temperature profiles inside yellow-green and green canopies on 29 May. Resistance thermometers were inserted at 10 cm above ground level

The most relevant temperature difference between green control and YG was found in the time between 11 a.m. and 3 p.m. Important consequences can arise from this experimental result when water transpirational loss is considered. SEIGNER [14] in fact calculated that when albedo increases from 0.25 to 0.40, the leaf surface temperature should be reduced by about  $3^{\circ}\text{C}$  and the evaporation by about 30%.

Chlorophyll contents and photosynthetic rate of flag leaf are shown in Tables 4 and 5, respectively.



**Table 4**  
*Chlorophyll a and b content*

	Wild type	YG
Chl-a (mg/g f w)	1.74	0.63
Chl-b (mg/dm <sup>2</sup> leaf surface)	3.22	1.06
Chl-b (mg/g f w)	0.62	0.18
Chl-b (mg/dm <sup>2</sup> leaf surface)	1.16	0.30
Chl-a		
Chl-b	2.8	3.5

**Table 5**  
*Photosynthetic and transpiration rates*

	T (°C)	Wild type	YG
Photosynthetic rate (mg CO <sub>2</sub> /dm <sup>2</sup> · hr)	23	21.4 (1.1)*	22.3 (1.3)
Photosynthetic rate (mg CO <sub>2</sub> /mg Chl · hr)	23	4.9 (0.3)	16.4 (1.0)
Transpiration rate (mg H <sub>2</sub> O/dm <sup>2</sup> · hr)	17	267 (34)	284 (26)
	23	566 (59)	675 (75)
	30	1079 (90)	1122 (119)

\* Standard errors in brackets

Experiments were made on flag leaf. The results are the mean value of six experiment. Light intensity in the photosynthetic assimilation chamber was 40 Klux and CO<sub>2</sub> concentration 320 µl/l.

Relative humidity 60%. T = air temperature in assimilation chamber.

Chl-a content of YG is a 1/3 with respect to wild type content both on the fresh weight and leaf area basis.

Higher deficiency of Chl-b is observed in YG. Therefore,  $\frac{\text{Chl-a}}{\text{Chl-b}}$  ratio is higher then in the wild type.

This is the most frequent case observed of the chlorophyll content in yellow and yellow-green mutants.

Carotenoid content of YG is a 1/2 with respect to wild type content. These phenomena can justify the higher transmission and reflection observed on YG. Although YG mutants exhibit a more reduced photosynthetic system (on chlorophyll basis), similar photosynthetic rates on area basis at saturated high intensity were found in both YG mutant and green control.

Higher values of this parameter were shown by the YG mutant on total chlorophyll basis (Table 5).

At saturating light intensity, the photosynthetic system of YG mutant seems more efficient on chlorophyll basis than that of the wild type.

No differences were observed in the transpirational rate, at different temperatures (Table 5). Further experiments [17] demonstrated similar photorespiration, RuDP carboxylase and RuDP oxygenase activity in both the wild type and the YG mutant.

Generally we can consider that net radiation absorbed by a canopy is dissipated as sensible and latent heat.

Lower absorption of net radiation by a canopy determines lower temperatures inside the same canopy and latent heat loss should be diminished.

Similar transpirational rates for green control and for yellow-green mutant were found; therefore, later transpirational loss should decrease because of lower temperatures inside the canopy.

Further physiological and agronomical investigations of crops on this cooler canopy will be necessary to verify the adaptability of yellow-green wheat variety to dry land agriculture.

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### References

- ALBERTE, R. S., HESKETH, J. D. and KIRBY, J. S. (1976): Comparison of photosynthetic activity and lamellar characteristics of virescent and normal green Peanut leaves. *Z. Pflanzenphysiol.*, **77**, 152–159.
- ALBERTE, R. S. and THORNER, J. P. (1974): The correlation between chlorophyll a/b ratio and proportions of chlorophyll-protein complexes in green plants. *Plant Physiol. (suppl.)*, **53**, 63.
- BILLING, N. D. and MORRIS, R. J. (1951): Reflection of visible and infrared radiation from leaves of different ecological groups. *Amer. J. Bot.*, 327–331.
- BENCI, J. F., AASE, J. K. and FERGUSON, A. M. (1973): Aerodynamic and energy balance comparisons between awned and non-awned barley. *Agron. J.*, 65.
- BENEDICT, C. R., MCKREE, K. J. and KOHEL, R. J. (1972): High photosynthetic rate of a chlorophyll mutant of cotton. *Plant Physiol.*, **49**, 968–971.
- CLEWELL, A. F. and SCHMID, G. H. (1969): Chlorophyll-deficient *Lespedeza procumbens*. *Planta*, **84**, 166–173.
- FERGUSON, M., COOPER, C. S., BROWN, J. M. and ESLICK, R. F. (1972): Effect of leaf color, chlorophyll concentration and temperature on photosynthetic rates of isogenic barley lines. *Agron. J.*, **64**, 671–673.
- FERGUSON, M., ESLICK, R. F. and AASE, J. K. (1973): Canopy temperatures of barley as influenced by morphological characteristics. *Agron. J.*, **65**, 425–428.
- GATES, D. M., KUGAN, H. J., SCHLETER, J. C. and WEIDER, V. R. (1965): Spectral properties plants. *Appl. Opt.*, **4**, 11–20.
- HIGHKIN, H. R., BOARDMAN, N. K. and GOODCHILD, D. J. (1969): Photosynthetic studies on a pea-mutant deficient in chlorophyll. *Plant Physiol.*, **44**, 1310–1320.
- JARVIS, P. G., CATSKY, J. (1971): General principles of gasometric methods and the main aspects of installation design. In Sestak, Z. et al. (eds): *Plant photosynthetic production manual of methods*. pp. 672–701. The Hague, Junk.
- KECK, R. W., DILLEY, R. A., ALLEN, C. F. and BIGGS, S. (1970): Chloroplast composition and structure differences in a soybean mutant. *Plant Physiol.*, **46**, 692–698.
- KOHEL, R. J. and BENEDICT, C. R. (1971): Description and CO<sub>2</sub> metabolism of aberrant and normal chloroplasts in variegated cotton, *Gossypium hirsutum* L. *Crop Science*, **11**, 486–488.
- SEIGNER, J. (1967): The effect of albedo on the evaporation rate. *Agr. Meteorol.*, **6**, 5–31.
- SINGH, B. P. (1978): Effect of leaf color on growth of spring barley. *Cer. Res. Communications*, **6**, 35–42.
- SESTAK, Z. (1971): Determination of chlorophyll a and b. In Sestak, Z. et al. (eds): *Plant photosynthetic production manual of methods*. pp. 672–701, The Hague, Junk.
- TRIOLO, L., CERVIGNI, T. and GIACOMELLI, M.: Photosynthetic characteristics of a yellow-green mutant of *durum* wheat. *Agrochimica* (in press).

# AMYLASE ACTIVITY IN VARIOUS MAIZE HYBRIDS

The enzyme system of the maize grain is one of the determinants of the quality of fodder. It is important to know the activity of enzymes decomposing the carbohydrates in maize, since a considerable part — some 55–80 per cent — of the maize grain is composed of carbohydrates, and a part of them is decomposed into easily soluble and available sugars by amylases active in the grain. Thus, the higher the amylase activity of the maize grain, the greater is the easily digestible, partly decomposed, absorptive carbohydrate content. It is also important to consider what amount of substrate is decomposed by the amylase, during preservation and storage, into simple sugars, as these are easier for animal digestion. The methods of preservation and storage, and the way they are applied, may cause essential changes in the activity of enzymes. Therefore, the enzyme activity measured can at the same time qualify these methods. Accordingly, we thought it important to find out:

a) what was the difference in active amylase content between maize varieties of different maturity groups;

b) how the method of preservation influenced the activity of enzymes;

c) what changes the above caused in the free reducing sugar content of the maize grain.

TOLLIER and GUILBOT (1971) studied the changes of amylase activity appearing in the maize grain during drying. According to their investigations, the amylase activity completely stops in maize grain dried at 140 °C. SALGÓ *et al.* (1978) pointed out that the amylase activity in cereals increased during germination. PÁRKÁNY *et al.* (1979) studying the amylase activity in various maize varieties, found substantial differences among the varieties.

A part of the techniques of measuring the amylase activity is based on the fact that, during the decomposition of starch or starch substrate, the amount of compounds released changes as a function of time, and from these changes, conclusions can be drawn on the enzyme activity (JOHNSON *et al.* 1964, PERTEN 1966). The measuring method used in our comparative study is based on the fact that the reducing final groups released from the surplus starch substrate in unit time react with dinitro-salicylic acid (DS). The quantity of coloured complex thus produced is measured with spectrophotometer (BERNFELD 1955, MORITA *et al.* 1975). For the purpose of determination, the maize grains were ground to flour and let through a screen of 80 mesh. The measured samples were extracted in 0.1 M acetate buffer (pH 5.4) for 30 minutes at 37 °C. The extract was treated with DS reagent at 100 °C, then cooled and photometried against a control at a wave-length of 540 nm.

The enzyme activity is expressed in amylase unit (AmU). The enzyme activity: AmU = 1  $\mu$ mol maltose produced/hour/g maize grain. The free reducing sugar content was measured directly from the extract of the sample.

Twelve maize varieties from three growing sites were analysed for amylase activity and free reducing sugar content, as well as for reducing sugar released with crystalline  $\alpha$ -amylase extracted from pig pancreas.

Table 1 shows the amylase activity in maize varieties of different maturity groups (FAO 200, FAO 300, FAO 400, FAO 500), when dried at 60 °C and preserved with a wet technique, respectively. The data of samples dried at 60 °C are mean values of samples obtained from three sites (Kaposvár, Székkutas, Debrecen). In the two early maize varieties (Pioneer hybrid 3978 SC and Pioneer hybrid 3950 SC), an average of  $4.80 \pm 0.85$  AmU was noted. In the medium early hybrids (Pioneer hybrid 3901 SC, Szegedi SC 390, Iris G 303 DC, Hibridor 212 MSC), the AmU was  $5.20 \pm 0.37$  on the average. The late varieties (MSC 394, OS 407 SK, MVSC 434, Pioneer hybrid 3732 SC, SC 3578, MVSC 550 WX) displayed an average AmU of  $5.73 \pm 1.36$ .

The forms of preservation greatly influence the activity of amylases. Table 1 also contains the amylase activity data of maizes preserved with a wet method (Pioneer hybrid



Table 1

*Amylase activity in maize hybrids preserved in different ways*1 amylase unit (AmU) = production of 1  $\mu$ mol maltose/hour/g maize

Preservation	Drying at 60 °C	Wet preservation
Variety (1980)		
Pioneer hybrid 3978 SC (FAO 200)	4.2	2.9
Pioneer hybrid 3950 SC (FAO 200)	5.4	—
	$\bar{x} = 4.80 \pm 0.85$	—
Pioneer hybrid 3901 SC (FAO 300)	4.7	1.6
Szegedi SC 390 (FAO 300)	5.6	2.7
Iris G 303 DC (FAO 300)	5.3	5.6
Hibridor 212 MSC (FAO 300)	5.2	—
	$\bar{x} = 5.20 \pm 0.37$	—
MSC 394 (FAO 400)	5.3	2.5
OS 407 SK (FAO 400)	7.3	2.6
MVSC 434 (FAO 400)	6.0	3.2
Pioneer hybrid 3732 SC (FAO 400)	6.8	—
SC 3578 (FAO 500)	3.4	3.4
MVSC 550 WX (FAO 500)	5.6	3.4
	$\bar{x} = 5.73 \pm 1.36$	$\bar{x} = 3.06 \pm 1.16$

Table 2

*Amylase activity in maizes dried at different temperatures*1 amylase unit (AmU) = production of 1  $\mu$ mol maltose/hour/g maize

Drying temperature	Drying at 90 °C	Drying at 130 °C
Variety (1979)		
Anjou SC 256 (FAO 200)	5.4	2.1
Szegedi SC 369 (FAO 300)	4.7	—
SC 3365 HL (FAO 300)	5.7	2.1
MVSC 580 (FAO 500)	3.8	1.7
	$\bar{x} = 4.90 \pm 0.85$	$\bar{x} = 1.96 \pm 0.23$
(1980)		
MV TC-296 (FAO 200)	4.2	0.4
SC 3385 HL (FAO 300)	4.7	2.1
Szegedi SC 369 (FAO 300)	6.3	2.9
SC 5443 (FAO 400)	3.8	1.2
	$\bar{x} = 4.75 \pm 1.09$	$\bar{x} = 1.65 \pm 1.08$

3978 SC, Pioneer hybrid 3901 SC, Szegedi SC 390, Iris G 303 DC, MSC 394, OS 407 SK, MVSC 434, SC 3578, MVSC 550 WX). In the nine samples preserved with a wet procedure, an average of  $3.06 \pm 1.16$  AmU was found. In Table 2 the amylase activity of maize varieties dried at 90 and 130 °C, respectively, is summed up. As seen from the values, drying at 130 °C significantly reduced the amylase activity in the samples examined; on the average, the value of AmU decreased from  $4.90 \pm 0.85$  to  $1.96 \pm 0.23$  in 1979, and from  $4.75 \pm 1.09$  to  $1.65 \pm 1.08$  in 1980, in response to drying at 130 °C.

Table 3 shows the percentage weight of free reducing sugars in maizes of different variety, maturity and preservation. The table reveals that Szegedi SC 390, Iris G 303 DC and Hibridor 212 MSC contained low quantities of free reducing sugars (0.32, 0.35 and 0.35 per cent, respectively). A high reducing sugar content was found in OS 407 SK, MVSC 550 WX, SC 3578 (0.77, 0.73 and 0.71 per cent, respectively). In maize hybrids dried at 60 °C the average free reducing sugar content was  $0.53 \pm 0.16\%$ .

**Table 3**  
*Free reducing sugar content in maize hybrids preserved in different ways*  
(in weight %)

Preservation	Drying at 60 °C	Wet preservation
Variety (1980)	(1980)	
Pioneer hybrid 3978 SC (FAO 200)	0.58	1.13
Pioneer hybrid 3950 SC (FAO 200)	0.70	—
Pioneer hybrid 3901 SC (FAO 300)	0.52	1.87
Szegedi SC 390 (FAO 300)	0.32	1.58
Iris G 303 DC (FAO 300)	0.35	1.00
Hibridor 212 MSC (FAO 300)	0.35	—
MSC 394 (FAO 400)	0.46	1.49
OS 407 SK (FAO 400)	0.77	1.65
MV SC 434 (FAO 400)	0.46	1.68
Pioneer hybrid 3732 SC (FAO 400)	0.48	—
SC 3578 (FAO 500)	0.71	1.94
MVSC 550 WX (FAO 500)	0.73	—
	$\bar{x} = 0.53 \pm 0.16$	$\bar{x} = 1.54 \pm 0.33$

The table also shows that in maizes stored wet, the free reducing sugar content increased threefold on the average. According to our measuring results, maizes stored wet contained  $1.54 \pm 0.33$  per cent free reducing sugar.

Drying at higher temperatures (90–130 °C) did not essentially affect the free reducing sugar content (Table 4).

Examinations were made to determine the amount of reducing sugars released with crystalline  $\alpha$ -amylase surplus, isolated from pig pancreas.

The amount of reducing sugars released by amylase from varieties of different maturity groups, obtained from different sites, is shown in Fig. 1 (Fig. 1). As seen in the figure, there is a 1.0–1.5 per cent difference in reducing sugar content releasable by  $\alpha$ -amylase among early, medium and late varieties in favour of the last. According to the results of our analyses made so far, medium and late varieties have a higher reducing sugar content. However, these correlations also need further investigations to be verified.

Table 4

*Free reducing sugar content in maize hybrids dried at various temperatures  
(in weight %)*

Variety	Drying temperature	
	90 °C	130 °C
Variety (1979)		
Anjou SC 256 (FAO 200)	0.43	0.52
Szegedi SC 369 (FAO 300)	0.48	0.34
SC 3365 HL (FAO 300)	0.57	0.43
MVSC 580 (FAO 500)	0.64	0.60
	$\bar{x} = 0.53 \pm 0.09$	$\bar{x} = 0.47 \pm 0.11$
(1980)		
MV TC 296 (FAO 200)	0.77	0.81
SC 3385 HL (FAO 300)	0.43	0.43
Szegedi SC 369 (FAO 300)	0.34	0.48
SC 5443 (FAO 400)	0.69	0.64
	$\bar{x} = 0.55 \pm 0.21$	$\bar{x} = 0.59 \pm 0.17$

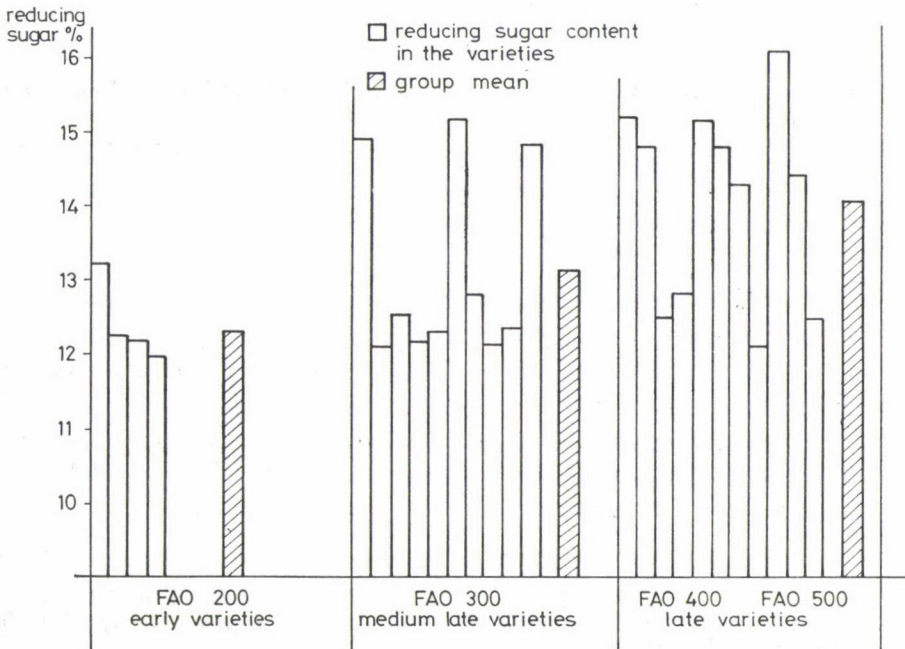


Fig. 1. Reducing sugar content released with crystalline  $\alpha$ -amylase



In our investigations the first aim was to apply a relatively quick method suitable for comparison, in measuring the amylase activity of maize samples, which can be then used as an analytical qualifying method, partly after literary descriptions and partly on the basis of revised works. The method of BERNFELD (1955) and MORITA (1975), modified by us, is considered suitable both for measuring the amylase activity and determining the reducing sugar content. On the basis of our measuring results, we have established that the amylase activity and reducing sugar content in maize are influenced by the crop's variety, site and year, in general, and by the time of maturing in particular. We intend to prove the effects of these factors, and the tendencies of the changes observed, with further analyses of a larger number of samples.

Preservation unambiguously has a significant effect on the parameters examined:

In comparison with samples dried at lower temperatures (60 °C), wet preservation reduced the activity of amylases to a lower- while drying at 130 °C to a higher extent (by 40 and 60 per cent on the average). Although the amylase activity decreased considerably on preservation, in consequence of a change in the value of pH caused by the lactic fermentation, a large amount of free reducing sugars is still released, supposedly through the decomposition of starch by microorganisms. According to our measuring results, in samples preserved wet the free reducing sugar content rose some three times higher than in the dried samples. Drying at higher temperatures, on the other hand, did not essentially influence the quantity of reducing sugars.

Both the decomposition of carbohydrates and the relatively high sugar content, produced in the course of their synthesis, are favourable from the point of view of digestibility and absorption. Wet preservation therefore provides a more valuable fodder for the animals, from the point of view of carbohydrate utilization. It has been established that the reducing sugar content releasable, by crystalline  $\alpha$ -amylase surpluses, is higher by an average of 1.5 per cent in the *later* than in the earlier varieties.

\*

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### References

- BERNFELD, P. (1955): *Methods in Enzymology* 1. Acad. Press, New York, p. 149.
- JOHNSON, G., LAMBERT, C., JOHNSON, D. K. and SUNDERWIRTH, S. G. (1964): Colorimetric determination of glucose and sucrose in plant material using a combination of enzymatic and chemical methods. *J. Agric. Food. Chem.*, **12**, 216.
- MORITA, Y., AIBARA, S., YAMOSHITA, H., YAGI, F., SAGANUMA, T. and HIROMI, K. (1975): Crystallization and preliminary X-ray investigation of soybean  $\beta$ -amylase. *J. Biochem.*, **77**, 343.
- PÁRKÁNY-GYÁRFÁS, A. and VÁMOS-VIGYÁZÓ, L. (1979): Determination of amylase activity in corn, using a chromogenic substrate. *Stroch/Stärke*, **10**, 328.
- PERTEN, H. (1966): A colorimetric method for the determination of  $\alpha$ -amylase activity. *Cereal Chem.*, **20**, 165.
- SALGÓ, A., ŐRSI, F. and SÜMEGHY, Z. (1978): Amilázok meghatározása "Contiflo" automatikus elemzőben (Determination of amylases in "Contiflo" automatic analyser). *Élelmiszervizsgálati Közlemények*, **25**, 173.
- TOLLIER, M. T. and GUILBOT, A. (1971): Caractéristiques de la fraction glucidique des échantillons de maïs grain. *Annales de Zootechnik*, **20**, 632.

## POTASSIUM FIXATION IN SANDY SOILS AS RELATED TO CLAY MINERALS

The previous studies showed that the Egyptian soils (Nile alluvials or marine sediments) have an abundance of potassium. The mineralogical studies showed a dominance of 2 : 1 layer silicate minerals. Increasing content of kaolinitic type minerals were associated with marine sediments rather than Nile alluvium. Potassium fertilizers are not commonly used for common field crops grown in Egypt. Vegetable and fruit crops however, seem to require potassium fertilizers for optimum yields.

The objective of this study is to identify the types of potassium fixing minerals and the potassium fixation capacity of two types of Egyptian soils and their subsize fractions (sand, silt and clay). This information is essential for nutritional purpose in particular that most of the newly reclaimed soils in Egypt are of lacustrine and marine origins.

*Fixed potassium:* VOLK (1934) defined the fixed potassium as that portion of added potassium which is not replaceable by hydrogen ions when the potassium treated soils is extracted by boiling 1 N  $\text{HNO}_3$  acid. KARIM and MALIK (1957) defined fixed potassium as, that portion of added potassium not exchanged with ammonium ions by extracting the soil with neutral normal ammonium acetate, or also not readily available to plants. PAGE and BAVER (1940) proposed the theory of fixation which suggested that fixation of potassium was possible due to the radius of hexagonal voids of the tetrahedral layer of the 2 : 1 clay minerals. The non hydrated potassium ions presumably move into these voids causing the structure to collapse. Only ions with diameter close to 2.8 Å are fixed to any considerable extent, the larger ions cannot enter the cavities, while ions that are too small cannot sufficiently stabilize the structure. LAMBERT (1950) studied fixation of potassium by clays saturated with different cations. The fixation was found to be very low when the clay was saturated with  $\text{H}^+$ ,  $\text{K}^+$  or  $\text{NH}_4^+$  but was very high when saturated with  $\text{Ca}^{2+}$  and especially with  $\text{Na}^+$ .

MORTLAND and GUSEKING (1951) studied the influence of the silicate ions on potassium fixation. They found that kaolinitic minerals when dried with potassium silicate fixed insignificant amounts of potassium. The illitic clay minerals fixed larger amounts of potassium which was not removed by boiling  $\text{HNO}_3$ . WEAR and WHITE (1951) studied potassium fixation in clay minerals as related to crystal structure. They concluded the following mechanism of fixation. When the exchange complex is saturated with potassium ions, they are held by attractive forces arising from both octahedral and tetrahedral charges at random distribution in the crystal structure. The beidellite and illite clays have more of the tetrahedral charges, which give rise to stronger attractive forces at clay surfaces. For illite it was found that a large amount of potassium was already fixed between the planes, and this situation decreased the exchange capacity, the hydration, and the swelling volume, and decreased subsequent fixation. RICH and BLACK (1964) showed that potassium fixation increased when  $\text{OH}^-$  arised. BARSHAD (1948, 1950 and 1954) showed that vermiculite can fix high amounts of potassium, hence it might lose about 60% of its exchange capacity. BARSHAD and KISHK (1968) showed that oxidation of octahedral  $\text{Fe}^{2+}$  to  $\text{Fe}^{3+}$  in soil vermiculite clays and boitites increased the potassium fixation capacity of vermiculite and increased the difficulty of replacing inter-layer potassium of biotite. They attributed the increase in the attractive forces between potassium ions and oxygen ions of the tetrahedral layer to tilting of the octahedral  $\text{OH}^-$  ion dipole from a perpendicular to an inclined (diagonal) position with respect to the cleavage plane.

MCLEAN and SIMON (1958) studied the potassium status of some Ohio soils as revealed by green house experiments. The exchangeable potassium reflected closely the potassium applied at all levels of application to the ten soils studied in the laboratory. Some added potassium was fixed against ammonium acetate in almost all soils even when kept moist, and this was largely beyond recovery even with boiling  $\text{nHNO}_3$ . Drying the soils in the



oven at 105 °C generally increased the exchangeable potassium at zero and low rates of potassium application, but it fixed increasing amounts at higher rates of application. Total acid extracted potassium was less affected by drying than were exchangeable forms. Their results showed an increase in the potassium released from non exchangeable form by the acid in the dried soils as higher rates of applied potassium were practiced. This may mean that the potassium fixed by drying is also largely recoverable by cropping, while that fixed under moist conditions and is not extractable by boiling  $\text{HNO}_3$ , is presumably not available to plants in short periods of cropping. In all soils, the amounts of potassium fixed in moist condition increased with the rate of application, but the fixed percentage of that applied decreased with higher rates of applications. The fine-textured soils caused a greater percentage of the total exchangeable potassium to be fixed as the rate of application increased while the medium, and coarse textured soils generally reached a maximum percentage of fixed potassium, and levelled off or dropped at the highest rate of application. The latter soils lost a higher percentage of potassium to fixation by drying than by moist fixation while the fine-textured soils generally lost as much by moist as by dry fixation. Various other points are also discussed which seem to indicate that as the laboratory data suggested, potassium fixed by drying was largely recoverable by cropping.

Potassium fixation is the transformation of exchangeable potassium into the non-exchangeable form by the migration of potassium ions into the crystal structure of micaceous minerals (illite and vermiculite) where they are strongly bound in vacant spaces originally occupied by potassium ions (WIKLANDER 1961).

In the fixed form, potassium exists between the non-exchangeable and exchangeable form, in which the ions are more tightly bound than the latter, although it is probably more available than the former. The fixed ions are not readily replaceable by neutral salts. DUTHION (1968) reported that the nature of clay fraction of the soils was most important in potassium fixation. This factor intervenes at the level of potassium fixation mechanism. It also affects the intensity of the process where fixation is zero with kaolinite, chlorite and micas, slight with montmorillonite, variable with illite according to their degree of alteration or weathering (potassium content) and strong with vermiculite. For montmorillonite, and after fixation occurs, the cation exchange capacity is found to be less by an amount corresponding to fixed potassium. The ions are now fixed at interfoliar sites which normally take part in the exchange. When dehydration occurs, the structure sheets come closer together and the adsorbed cations lose their attached molecules of water. The outer layers of sheets in clay minerals are composed of assays of oxygen atoms which have between them hexagonal cavities of 2.8 Å diameter. The potassium ions, which when dehydrated have a diameter of 2.66 Å, can fit into those cavities and, thus, are trapped between two adjacent sheets. The configuration formed is practically stable and is resistant to rehydration.

Other factors of importance in the process of potassium fixation may be:

- a) adsorbed cations,
- b) associated anion,
- c) soil pH,
- d) organic matter,
- e) lime effect,
- f) temperature, and
- g) size fraction.

ROTH *et al.* (1968 and 1969) showed that the cation exchange increased 10–60% as a result of deferration of micaceous vermiculites and soils. Reoxidation of the deferrated samples restored the  $\text{Fe}^{3+}$  content to approximately the original value but the CEC was not affected. The reversible change in valence of the structural Fe without an equivalent change in CEC



was attributed to deprotonation-protonation of the structure ( $\text{OH}^- \rightleftharpoons \text{O}^{2-}$ ), simultaneous with the oxidation reduction of the structural ( $\text{Fe}^{2+} \rightleftharpoons \text{Fe}^{3+}$ ) in the phyllosilicate layer. The interplanar charge density of phyllosilicates affects the release and fixation of a cation and the tendency of these minerals to expand. BARSHAD (1948) estimated that boitite minerals which had more than 158 m.e/100 g of inter-layer charge, were not subject to expansion.

Factors that affect the rate of potassium release to weathering solutions are; temperature, pH, total surface charge, concentration of different ions in the solution, perfection of the structure, the Fe/Mg of the mineral structure and the type of octahedral cations (di or tri octahedral).

SCHULTE and COREY (1965) showed that the critical value of potassium ions in the exterior solution that prevents the release of inter-layer potassium may be as little as 0.025 m.e/l. SAWHNEY (1969) showed that sorption of calcium or potassium by calcium saturated vermiculite produced regularly interstratified mica-vermiculite layer sequence, while montmorillonite formed random interstratification which was related to the layer charge density of the minerals.

The soil samples used in this study are from the north-western region of Egypt.

Al-Nahda represents the southern margin of the lacustrine deposits of lake Mariut, and a transitional area to the marine deposits of the western desert of Egypt. This project is located east of the main Alexandria—Cairo desert road between km 34 and 40. Only the lacustrine deposits were used in this study. These deposits are highly saline, contain relatively a high percentage of secondary  $\text{CaCO}_3$  and  $\text{CaSO}_4$  plus an abundance of shells, and are of clay textures.

The northern Tahreer project is located just south of Al-Nahda. The soils here are formed from the coastal and desert marine sediments and are highly calcareous, slightly saline, and of medium to coarse texture.

The samples were taken from soil profiles, air dried, passed through a 20 mesh sieve, stored, and used for study. The clay, silt, and sand fractions were obtained from dispersed samples by sodium-hexametaphosphate and shaking for at least 72 hours. The clay fraction was separated by siphoning the suspended clay in 1 liter cylinders after 24 hours. The coarse, medium and fine sands were sieved through a 59 mesh screen. The very fine sand and silt fraction were collected together. The clay and silt fractions were kept wet while the sand was air dried. It should be noted that the carbonates were not removed from the sample or any of the fractions during this study.

The following analyses were made on the soil samples and all other fractions if possible.

1. Mechanical analysis, was carried out by the Hydrometer method.
2. Total salts, in the saturated extract were measured by electric-conductivity (Beckman conductivity meter, type CD M<sub>2</sub>d) (RICHARDS 1954).
3. Total carbonates, as  $\text{CaCO}_3$  equivalent was determined using a simple calcimeter.
4. Cation exchange capacity; suitable weights of soil, or other soil size fractions were sodium-saturated by 1 N Na Ac, washed till salt-free by distilled water and ethyl alcohol plus acetone, then sodium was extracted by 1 N ammonium acetate at pH 8.2 (BLACK 1965).
5. Potassium fixation capacity (PFC); samples were K-saturated by three washings of 25 ml of 0.1 N KCl followed by two washings with alcohol, then samples were heated for 24 hours at 110 °C. Non-fixed potassium (released K) was extracted in a normal ammonium acetate (EL-ATTAR 1970).  $\text{PFC} = \text{CEC} - \text{Released K}$ .
6. Amorphous silica and alumina were dissolved in boiling 0.5 N NaOH solution from samples before and after dehydroxylation at 600 °C for 6 hours (JACKSON 1965).
7. Deferrated iron by Na-dithionite in a buffered solution of bicarbonate and citrate sodium solutions was determined in dehydroxylated samples by colorimeter (KRISHNA MURTI *et al.* 1966).

8. The amorphous material percentage was calculated as proposed by HASHIMOTO and JACKSON (1960) as follows:

$$\text{Am. \%} = \frac{\text{SiO}_2 \% + \text{Al}_2\text{O}_3 \%}{0.9}$$

where:  $\text{SiO}_2\%$  and  $\text{Al}_2\text{O}_3\%$  are the silica and alumina percentage in the samples before dehydroxylation. The amorphous material contains 10%  $\text{H}_2\text{O}$ .

9. The kaolinitic type mineral content is calculated from the differences between silica and alumina percentage in samples after and before dehydroxylation.

$$\begin{aligned}\text{Kaolinite \%} &= \frac{\Delta \text{SiO}_2 \%}{0.456} \\ \text{or} &= \frac{\Delta \text{Al}_2\text{O}_3 \%}{0.395}\end{aligned}$$

or = the average of the above two values;

where:

kaolinite contains 45.6% of  $\text{SiO}_2$  and 39.5% of  $\text{Al}_2\text{O}_3$ ,

$\Delta \text{SiO}_2\%$  =  $\text{SiO}_2\%$  after dehydroxylation -  $\text{SiO}_2\%$  before dehydroxylation, and

$\Delta \text{Al}_2\text{O}_3\%$  =  $\text{Al}_2\text{O}_3\%$  difference as the above.

10. The montmorillonitic and vermiculitic type mineral content is estimated on the bases of the change in the cation exchange capacity values after different specified treatments of samples (ALEXIADES and JACKSON 1965).

$$a) \text{ Vermiculite \%} = \frac{\text{CEC} - \text{CEC}/\text{NH}_4}{150} \cdot 100;$$

where:

CEC = cation exchange capacity of Na-saturated samples,

$\text{CEC}/\text{NH}_4$  = cation exchange capacity of K-saturated samples after heating at 110 °C for 24 hours and extracted by  $\text{NH}_4$  solution,

150 = the average interlayer charge of vermiculite.

$$b) \text{ Montmorillonite \%} = \frac{\text{CEC}/\text{NH}_4}{105} \cdot 100;$$

where:

$\text{CEC}/\text{NH}_4$  = the same as above and it represents the surface charge of all minerals other than vermiculitic minerals,

105 = average surface charge of montmorillonitic minerals.

Modified methods of calculation of minerals are used in this study. These modifications are suitable for a mineralogical estimation to serve the purpose of this study. An accurate determination of the mineralogical composition requires the removal of all cemented materials which may be present in the samples, such as calcium carbonate, which opposes the objectives of this study.

**Potassium fixation capacity (PFC):** Fixed potassium is determined on samples which are previously potassium saturated and heated at 110 °C for 24 hours to perform maximum potassium fixation.

Al-Nahda soil fixes between 5.4 and 12.8 m.e/100 g while Al-Tahreer soil fixes only from 1.8 to 5.3 m.e/100 g. Both soils are of low capacity to fix potassium, but Al-Nahda soil fixes as twice as Al-Tahreer soil (Table 1).

Table 1

*The water soluble, exchangeable, available potassium and the potassium fixation capacity (PFC) and the cation exchange capacity (CEC) of Al-Nahda and Al-Tahreer soils*

Samples depth, cm	Potassium			PFC		CEC
	in H <sub>2</sub> O	in NH <sub>4</sub> Ac	in HNO <sub>3</sub>	Released-K	Fixed-K	
	m.e./100 g					
<i>Al-Nahda soil</i>						
0- 5	1.08	1.32	4.88	12.0	6.0	18.0
5- 15	1.12	1.98	5.92	10.6	9.0	19.6
15- 35	1.08	2.32	7.20	15.2	12.8	28.0
35- 65	1.04	2.36	5.68	18.8	5.4	24.0
Mean	1.07	2.21	5.67		8.2	24.1
<i>Al-Tahreer soil</i>						
0- 30	0.140	1.26	2.96	10.6	2.6	13.2
30- 60	0.098	0.77	2.16	12.0	5.0	17.0
60- 80	0.108	0.59	2.04	10.2	5.3	15.5
80-110	0.050	0.77	2.76	10.2	2.8	13.0
110-150	0.102	0.59	2.04	15.2	1.8	17.0
Mean	0.100	0.80	2.40		3.5	15.1

Table 2

*The exchangeable, available, released potassium and the potassium fixation capacity (PFC) and the cation exchange capacity of Al-Nahda and Al-Tahreer silty fractions*

Samples depth, cm	Potassium		PFC		CEC
	in NH <sub>4</sub> Ac	in HNO <sub>3</sub>	Released-K	Fixed-K	
	m.e./100 g				
<i>Al-Nahda</i>					
0- 5	3.3	8.75	37.96	0.47	38.43
5- 15	4.6	6.92	34.20	0.42	34.62
15- 35	6.0	12.02	15.61	2.01	17.62
35- 65	8.5	13.34	32.48	6.79	39.26
Mean	6.7	11.60		3.80	31.40
<i>Al-Tahreer</i>					
0- 30	6.22	6.0	23.29	0.25	25.52
30- 60	0.46	1.5	7.66	0.51	8.17
60- 80	0.34	2.7	4.16	1.03	5.16
80-110	0.30	1.2	4.13	1.03	5.16
110-150	0.56	1.4	4.77	1.26	6.02
Mean	1.60	2.6		1.20	10.00



**Table 3**

*The available potassium, the potassium fixation capacity and the cation exchange capacity of Al-Nahda and Al-Tahreer sandy fractions*

Samples depth, cm	Potassium in HNO <sub>3</sub>	PFC		CEC
		Released-K	Fixed-K	
		me/100 g		
<i>Al-Nahda</i>				
0– 5	2.8	16.26	1.69	17.95
5– 15	3.2	27.91	2.74	30.74
15– 35	3.3	23.99	0.75	24.74
35– 65	2.9	17.39	8.13	25.52
Mean	3.1		4.50	25.50
<i>Al-Tahreer</i>				
0– 30	1.60	5.35	1.15	6.50
30– 60	1.76	2.95	2.66	5.61
60– 80	1.60	4.32	2.50	6.82
80–110	1.60	3.13	3.63	6.76
110–150	1.72	3.10	4.05	7.16
Mean	1.65		2.80	6.30

**Table 4**

*The cation exchange capacity, released and fixed potassium (PFC) after heating at 110 °C overnight of Al-Nahda, and Al-Tahreer soil clays*

Samples depth, cm	Clay %	CEC	PFC	
			Released-K	Fixed-K
			me/100 g	
<i>Al-Nahda</i>				
0- 5	40	58.0	31.6	26.4
5- 15	40	64.0	32.0	32.0
15- 35	60	60.0	35.2	24.8
35- 65	50	42.0	25.0	17.0
Mean	50.8	52.1		22.5
<i>Al-Tahreer</i>				
0- 30	30	46.00	22.6	23.40
30- 60	20	40.82	19.0	21.82
60- 80	20	50.00	20.0	30.00
80-110	25	60.00	19.8	40.20
110-150	50	52.00	20.4	31.60
Mean	29	50.00		29.20

Table 5

*The particle size distribution and the total carbonate equivalents in soils and their subsize fractions*

Samples depth, cm	Total soil carbonate, %	Size fractions and their carbonate					
		Sandy		Silty		Clay	
		Sand%	CaCO <sub>3</sub> %	Silt%	CaCO <sub>3</sub> %	Clay%	CaCO <sub>3</sub> %
<i>Al-Nahda</i>							
0– 5	14.6	45	21.4	15	20.6	40	7.6
5– 15	20.0	50	17.2	10	22.3	40	8.7
15– 35	20.5	30	20.6	10	29.4	60	9.7
35– 65	21.5	40	34.4	10	15.5	50	22.3
Mean	20.3	39	26.5	10.4	21.0	50.8	15.2
<i>Al-Tahreer</i>							
0– 30	24.4	45	12.6	25	26.6	30	18.0
30– 60	33.0	40	7.6	40	42.5	20	24.5
60– 80	43.7	40	7.5	40	34.0	20	17.5
80–110	31.3	40	18.9	35	34.0	25	19.5
110–150	27.0	35	8.8	15	27.3	50	20.1
Mean	32.0	40	11.0	31	33.0	29	20.0

Table 6

*The percentage of vermiculitic type minerals in the soils and their subsize fractions*

Samples depth, cm	Soil			
	Vr. %			
	Soil	Sandy	Silty	Clay
<i>Al-Nahda</i>				
0- 5	3.89	1.10	0.31	17.14
5- 15	5.84	1.78	0.27	20.78
15- 35	8.31	0.49	1.30	16.11
35- 65	3.51	5.28	4.41	11.04
Mean	5.40	2.90	2.50	14.50
<i>Al-Tahreer</i>				
0- 30	1.69	0.75	1.46	15.19
30- 60	3.25	1.73	0.33	14.17
60- 80	3.44	1.62	0.67	19.48
80-110	1.82	2.36	0.67	26.10
110-150	1.17	2.63	0.82	20.52
Mean	2.70	1.80	0.80	19.10

**Table 7**  
*The percentage of montmorillonitic type minerals  
 in the soils and their subsize fractions*

Samples depth, cm	Soil	Sandy	Silty	Clay
	Mont. %			
<i>Al-Nahda</i>				
0- 5	11.43	15.49	36.15	30.10
5- 15	10.09	26.58	32.57	30.48
15- 35	14.48	22.85	14.87	33.52
35- 65	17.90	16.56	30.94	23.84
Mean	15.10	20.00	27.00	28.30
<i>Al-Tahreer</i>				
0- 30	10.09	5.10	22.18	21.52
30- 60	11.43	2.81	7.30	18.10
60- 80	9.71	4.11	3.96	19.05
80-110	9.71	2.98	3.94	18.86
110-150	14.48	3.86	4.54	19.43
Mean	11.10	3.80	8.40	19.40

The fixed potassium represents 23 to 46% and 11 to 34% of the exchange capacity of the two soils, respectively. This result may indicate that the vermiculitic type minerals represent 20 to 40% of the montmorillonitic type non-fixing minerals in Al-Nahda, but only 10 to 30% of the 2 : 1 layer silicates in Al-Tahreer (Table 6, 7).

The vermiculitic and montmorillonitic type minerals are estimated on the basis of the change in the cation exchange capacities upon certain specific treatments. The potassium fixation by vermiculitic minerals is assumed upon heating a potassium saturated sample at 110 °C for 24 hours. A reduction in the CEC, equivalent to the fixed potassium represents the vermiculite inter-layer charge. The montmorillonitic type minerals charge is equivalent to the residual CEC after potassium fixation.

The estimated vermiculitic minerals are 5.4% and 2.7% for Al-Nahda and Al-Tahreer soils, respectively (Table 6). The montmorillonitic minerals are 15.1% and 11.1% for the two soils, respectively (Table 7).

The clay fractions fixed 17.0 to 32.0 m.e/100 g and 22.0 to 40.0 m.e/100 g for Al-Nahda and Al-Tahreer clays, respectively (Table 4). These values represent 40 to 50% and 50 to 67% of the cation exchange capacity of these clays, respectively.

The vermiculitic clay content mean values are 14.5% and 19% while the montmorillonitic content mean values are 28.3% and 19% for the two soil clays, respectively.

Al-Tahreer clays contain equal amounts of montmorillonitic and vermiculitic minerals, but Al-Nahda clays contain twice the amount of montmorillonite as the amount of vermiculitic mineral. This finding accords with both the cation exchange and fixation capacities of the two types of clay fractions (Table 4). The carbonate content (Table 5) in Al-Tahreer clays is also higher than in Al-Nahda clays, which may cause a reduction in their exchange and fixation capacities.

The silty fraction (very fine sand, plus silt) fixed potassium in the range of 0.4 to 6.8 and 0.5 to 2.3 (mean 3.8 and 1.2) m.e/100 g for Al-Nahda and Al-Tahreer silts, respectively (Table 2). This finding accords with the carbonate content in this fraction. The surface layers



fixed low amounts compared with subsoil samples, because of high montmorillonitic content. The exchange capacity of Al-Nahda silt is relatively high. These values of exchange capacity represent 200% of the value of soil exchange capacity (Table 1). This result may mean an abundance of montmorillonitic type minerals. Also these high values of CEC of the silty fraction (in this study) may also be due to the presence of aggregates of silt-clay particles in this fraction. Exchange capacity of high values (200% of the cation exchange capacity of the soil) are found for the silt of the surface layer of Al-Tahreer soil, however, the subsoil silt showed cation exchange capacity values of about 50% of the soil itself. The surface layer has low carbonate content. It may also be affected by cultivation practices. The vermiculitic type minerals are estimated to be low (Al-Nahda 2.5%), or very low (Al-Tahreer 0.8%) in the silty fractions (Table 6).

The vermiculite in the silty fraction is lower than the sand and soil vermiculite content. The silt fractions on the other hand, contain higher percentage of montmorillonite minerals (27% and 8% for the two silts respectively). The presence of this type of mineral explains the high exchange capacity values of the silts, especially that of Al-Nahda. These results also prove that the montmorillonitic minerals in the silt fractions have low fixation capacity. A remarkable result of this study is that the montmorillonitic minerals percentage of the silt and clay fractions are about equal for Al-Nahda lacustrine soils (27 and 28% in both fractions, respectively). This result holds true only for the surface layer of Al-Tahreer; the subsoil layers contain high montmorillonite and low vermiculite. The carbonate content averaged 22 and 33% of the silty fraction of Al-Nahda and Al-Tahreer, respectively (Table 5). This about equals the carbonate percentage throughout the whole soil. The high content of carbonate in the silt fraction especially in Al-Tahreer (33%) may serve to explain the low values of cation exchange capacity and potassium fixation capacity of this silty fraction. It should be observed here that the carbonate effect is assumed to be larger in Al-Tahreer than Al-Nahda, which may be due to a difference between the nature and distribution of the carbonate minerals in the two silty fractions, hence in the two soil types. The carbonate activity and effect may be of different values because of the difference in their origin; they are mainly of aragonitic type (shells) in Al-Nahda lacustrine soil (El-Attar, 1970). Further studies may be required to identify the activity values and the distribution of the two types of carbonate present in these soils.

The sandy fraction, similar to the silty, fixed low amounts of potassium; however, the fixation capacity of the sand fraction is much higher than that of the silt fraction. Al-Tahreer sand fraction fixed potassium as much as 18 to 57% of its exchange capacity. The exchange capacity of Al-Nahda sand fraction is much higher than that of Al-Tahreer sand fraction (Table 3). These results are explained by the higher content of montmorillonitic-type minerals in Al-Nahda than in Al-Tahreer sand fraction (Table 7). The sandy fraction in this study may contain aggregates of silty-clay particles.

Vermiculitic minerals in the two sand fractions are about equal; 2.9 and 1.8%. The carbonate content of Al-Nahda sand fraction (mean value 26.5%) is slightly higher than the soil carbonate content (mean value 20.3%). Al-Tahreer sand, on the other hand, contains about 11% of carbonate and the soil contains 32% on the average. The carbonate content in the two sands is not in accordance with their exchange or fixation capacities. This result shows that the nature of carbonate content differs in the two soils. It also shows that the interference of carbonate with the exchange or fixation differs according to different size fractions. We may recall that the carbonate content of the silty and clay fractions are in better agreement with the last two parameters.

The kaolinitic type minerals are low in both soils, but being of larger percentage in Al-Tahreer than Al-Nahda (Table 8). The amorphous material content is about equal in both soils and subsize fractions (Table 9).

Table 8

*The percentage of kaolinitic type minerals in the soils  
and their subsize fractions*

Depth, cm	Soils			Sandy			Silty			Clay		
	A	B	Ave	A	B	Ave	A	B	Ave	A	B	Ave
<i>Al-Nahda</i>												
0- 5	1.4	0.9	1.2	1.4	1.3	1.3	6.3	3.9	5.1	8.1	3.6	5.8
5- 15	1.4	1.3	1.4	1.8	1.6	1.7	5.8	3.9	4.8	8.1	3.6	5.8
15- 35	2.3	1.3	1.8	2.1	1.5	1.8	3.5	3.6	3.5	6.9	5.9	6.4
35- 65	1.8	1.3	1.6	1.4	1.5	1.5	4.6	3.6	4.1	11.5	7.1	9.3
Mean			1.6			1.6			4.4			6.8
<i>Al-Tahreer</i>												
0- 30	3.2	1.5	2.3	1.2	1.6	1.4	5.8	3.9	4.8	8.1	7.1	7.6
30- 60	2.8	1.4	2.1	0.7	0.8	0.7	5.8	3.8	4.8	11.5	6.8	9.1
60- 80	3.5	1.5	2.5	0.7	0.8	0.7	5.8	3.8	4.8	11.5	6.8	9.1
80-100	2.5	0.9	1.7	0.9	0.9	0.9	4.0	3.2	3.6	11.5	5.7	8.6
110-150	4.6	1.4	3.0	0.9	0.9	0.9	4.0	4.6	4.3	15.0	7.3	11.1
Mean			2.1			0.95			4.5			9.0

$$A = \frac{\text{SiO}_2 \text{ } \%/}{0.465}$$

$$B = \frac{\text{Al}_2\text{O}_3 \text{ } \%/}{0.395}$$

Table 9

*The percentage of amorphous material in the soils  
and their subsize fractions*

Sample depth, cm	Soil	Sandy	Silty	Clay
amor. %				
<i>Al-Nahda</i>				
0- 5	0.38	0.52	1.66	3.90
5- 15	0.56	0.90	2.12	3.90
15- 35	0.38	0.85	1.66	3.67
35- 65	0.53	0.90	2.25	3.53
Mean	0.50	0.80	1.90	3.70
<i>Al-Tahreer</i>				
0- 30	0.62	0.85	2.48	4.74
30- 60	0.62	0.85	2.25	1.90
60- 80	0.62	0.85	2.25	3.98
80-110	0.53	0.61	1.98	5.29
110-150	0.56	0.73	1.30	5.29
Mean	0.60	0.80	2.00	4.20

The clay mineralogical composition of the two soils and their sand, silty and clay size fractions are included in this study. The mean values of the vermiculite type minerals (potassium fixing mineral) are 5.4 and 2.7%, and 2.9 and 1.8%, 2.5 and 0.8%, and 14.5 and 19.1 for the soil and sandy, silty, and clay fractions from Al-Nahda and Al-Tahreer, respectively. The montmorillonite type minerals represented 15.1 and 11.1%, and 20.0 and 3.8%, 27.0 and 8.4%, and 28.3 and 19.4% of the soil and sandy, silty and clay fractions from Al-Nahda and Al-Tahreer, respectively. The kaolinite type minerals represented 1.6 and 2.1%, and 1.6 and 1.0%, 4.4 and 4.5%, and 6.8 and 9.0% of the soil and sandy, silty and clay fractions from the two soil types, respectively. The amorphous material contents are 0.5 and 0.6%, and 0.8 and 0.8%, and 1.9, 2.0% and 3.7 and 4.2% in the soil and sand, silty and clay fractions of the two soil samples, respectively. The amorphous material difference between the two soils and their size fractions is not very significant. The montmorillonite values are higher for Al-Nahda than Al-Tahreer while the kaolinite values are reversed. The vermiculite values are higher in Al-Nahda samples except in the clay fraction where Al-Tahreer gives a higher value.

The potassium bearing minerals, however, may fix ammonium ion, therefore, the potassium in an ammonium extract will not be a satisfactory parameter for potassium availability. Also the acid extract will release larger amount of non-exchangeable potassium.

The potassium fixing capacity of the sample is proportional to its vermiculitic type minerals content. This is especially clear for the clay fraction of Al-Tahreer soil. This result suggests the importance of the mineral complex identification for potassium assessments.

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### References

- BARSHAD, I. (1948): Vermiculite and its relation to biotite as revealed by base exchange reaction, X-ray analysis, differential thermal curves and water content. *Amer. Mineral*, **33**, 655-678.
- BARSHAD, I. (1950): The effect of the inter-layer cations on the expansion of the mica type of crystal lattice. *Amer. Mineral*, **35**, 223.
- BARSHAD, I. (1954): Cation exchange in micaceous minerals. II. Replaceability of  $\text{NH}_4^+$  and  $\text{K}^+$  from Vermiculite, Biotite, and Montmorillonite. *Soil Sci.*, **78**, 57-76.
- BARSHAD, I. and KISHK, F. M. (1968): Oxidation of ferrous ion in vermiculite and biotite alters fixation and replaceability of potassium. *Science*, **162**, 1401-1402.
- DUTHION, C. (1968): Potassium in the soil. *Potash Review*, 4/43.
- KARIM, A. Q. M. B. and MALIK, M. A. (1957): Potassium fixation in East Pakistan. *Soil Sci.*, **83**, 229.
- LAMBERT, W. (1950): Fixation of potassium by clays saturated with different cations. *Soil Sci.*, **69**, 261-268.
- MORTLAND, M. M. and GUSEKING, J. E. (1951): Influence of the silicate ion on K-Fixation. *Soil Sci.*, **71**, 381-358.
- MCLEAN, E. O. and SIMON, R. H. (1958): Potassium status of some Ohio soils as revealed by green house and laboratory studies. *Soil Sci.*, **85**, 324.
- PAGE, J. B. and BAVER, L. D. (1940): Ionic size in relation to fixation by colloidal clay. *Soil Sci. Soc. Amer. Proc.*, **4**, 150.
- RICH, C. I. and BLACK, R. W. (1964): Potassium exchange as affected by cation size, pH and minerals structure. *Soil Sci.*, **97**, 384-390.
- ROTH, C. B., JACKSON, M. L., LOTSE, E. G. and SYERS, J. K. (1968): Ferrous-Ferric ratio and CEC changes on deferration of weathered micaceous vermiculite. *Israel Jour. of Chem.*, **6**, 261-273.
- ROTH, C. B., JACKSON, M. L. and SYERS, J. K. (1969): Deferration effect on structural ferrous, ferric ions ratio and CEC of vermiculites and soils. *Clays and Clay Minerals*, **17**, 253-264.



- SAWHNEY, B. L. (1969): Regularity of interstratification as affected by charge density in layer silicates. *Soil Sci. Soc. Amer. Proc.*, **33**, 42.
- SCHULTE, E. E. and COREY, R. B. (1965): Extraction of K from soils with sodium tetraphenylboron. *Soil Sci. Soc. Amer. Proc.*, **29**, 33-36.
- VOLK, M. J. (1934): The fixation of potash in difficultly available forms in soils. *Soil Sci.*, **37**, 267-287.
- WEAR, J. I. and WHITE, J. L. (1951): Potassium fixation in clay minerals as related to crystal structure. *Soil Sci.*, **71**, 1-14.
- WIKLANDER, I. (1961): Potassium in the cultivated soils in the province of Skoane. *Potash Review Subj.*, 5/18, 1-19.

#### EFFECT OF SOIL MOISTURE AND AERATION ON THE EMERGENCE OF RAW AND PELLETED SUGAR BEET SEED

The problems of sugar beet stand establishment have been studied very extensively but mainly from standpoint of possible adverse effects of fungal diseases and pests. Information about the effect of soil conditions on emergence is relatively scarce. Only one paper up to now (STEHLIK and NEUWIRTH 1928) deals with stand establishment as a comprehensive problem and treats the ecological soil conditions as the most important factor affecting emergence and seedling survival. The most critical period is usually supposed to be from the time of seed swelling to the phase of four true leaves. Fungi and pests can destroy very sensitive germs or seedlings, but it is primarily the suitable ecological conditions that enable the seed to germinate and emerge. The importance of ecological factors is asserted by WILLEY (1971). He came to the conclusion that the most important factor was the effect of the year, followed by the effect of the site. The effect of genetic factors on the stands was of less importance.

It has been established many times (see VEVERKA 1982) that sugar beet seed is very sensitive to high substrate moisture in the laboratory germination tests, and similar sensitivity may occur in the field (PERRY 1973). Germination of sugar beet decrease in wet soil more intensively than that of other plant species (FISCHNICH and GRIMM-THIELEBEIN 1959, HEYDECKER and GULLIVER 1972). LONGDEN (1973) showed the decline of sugar beet emergence to be affected by moisture in three different soil types, the least emergence occurring in the heaviest soil.

The effect of excessive or inadequate moisture content of the soil on germination and emergence was studied by AURA (1975) and STOUT, SNYDER and CARLETON (1959). Different seedlots are not equally sensitive to water excess. Relatively less sensitive seedlots absorb water more slowly than the sensitive ones (SNYDER and ZIELKE 1973). Pelleted seed is even more sensitive to water excess (VANSTALLEN 1971, AURA 1975, VEVERKA *et al.* 1979, VEVERKA 1982), but this may not be reflected in the field trials (HIBBERT, THOMSON and WOODWARK 1972). VANBREMEERSCH (1972) derived theoretical curves of the effect of the amount of pelleting material on germination. The layers of pelleting material serve as a barrier to oxygen diffusion and an increase in pellet size reduces seedling emergence. Very little information is yet available on the effect of low soil moisture on sugar beet emergence (ROSS and HEGARTHY 1979). According to HUNTER and ERICKSON (1952) sugar beet does not germinate if the soil water potencial is below  $-3.5$  atm, as distinct from soya ( $-6.6$  atm.) or maize ( $-12.5$  atm.). The germination of different cultivars was inhibited to the same degree by low water potencial (AURA 1975).

The basic principles of water and oxygen availability in the soil were published by CURRIE (1961), WENGEL (1966), GARDNER (1968) and BLACK (1968). A review of the effects of water stress on seed germination is given by HEGARTHY (1978).

Recently we have studied germination of raw and pelleted seeds under laboratory con-

ditions and derived a theoretical model of the effects of water and oxygen availability on the germination of sugar beet seed. In this paper we shall demonstrate the effect of soil moisture and aeration on the emergence of raw and pelleted seed.

Seed samples of monogerm var. "Monohybrid", raw and pelleted in UNS material (wood dust + fly ash) were the same as recently used (VEVERKA 1982). Fungal diseases were checked by seed treatment with Heryl (80% thiram) 12 g per 1 kg of seed and by damp sterilization of the soil. Trials were done in four replications in 10×10 cm plastic pots, filled with 0.6 kg dry soil sieved through 2 mm mesh. The soil was moistened to the capillary capacity, allowed to dry to the lowest moisture used in tests, taken out, mixed, returned to the pots, sown 2 cm deep, firmed (5 kg per 100 cm<sup>2</sup> for 2 minutes) and adjusted to the various moistures. On each of the following days, the moisture was checked by weight and adjusted by overhead watering, at a constant temperature range of 18–20 °C.

The maximum capillary capacity was previously checked by cylinders (100 cm<sup>3</sup>) after 2 hours redistribution into filtering paper. Aeration was calculated in relation to soil moisture. Soil conditions: porosity 56.2%, capillary capacity 39.6 (W/W), minimal air capacity 16.6%, specific density 2.56 g · cm<sup>-1</sup>, soil particles: >0.25 = 5.9%, 0.25–0.05 mm = 17.3%, 0.05–0.01 = 35.7%, <0.01 = 41.1% (0.001 = 22.5%).

The rate of emergence of pelleted seed (Fig. 1) was much lower than that of raw seed, except at a moisture of 12.5%. No emergence occurred at a moisture of 10%, and emergence at 15% moisture was significantly lower than in the raw seed. At higher moisture the emergence of pelleted seed was negligible, unlike the raw seed which did well even at 30% moisture. Raw seed reached the highest emergence rate of 66%, but the pelleted one reached 42.0% only.

The final emergence (Fig. 2) also shows the different response of raw and pelleted seeds to the various soil moisture content. The moisture of 7.5% was also too low for raw seed, which performed well in the soil moisture range of 10–25%; but at 30% moisture emergence decreased to 52.2%, at the maximum capillary capacity of 7.5%. Pelleted seed did not emerge at the soil moisture of 7.5%, and at 10% moisture the emergence was very low (14.6%). Optimum emergence was achieved at a 12.5–15% moisture content. At higher soil moisture content, emergence of pelleted seed was very low.

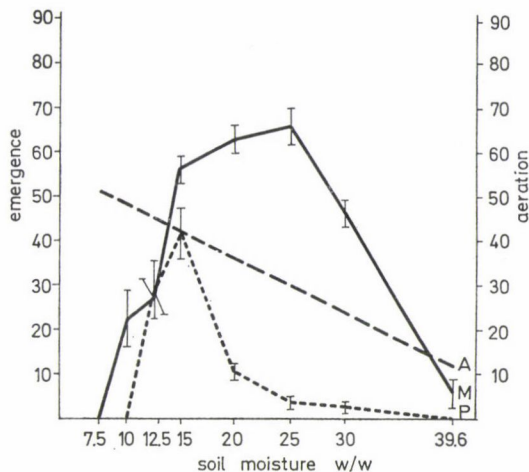


Fig. 1. Effect of soil moisture and aeration on the rate of emergence of raw and pelleted sugar beet seed. M ——— raw seed var. Monohybrid; P ——— pelleted seed; A = aeration (evaluation of the trial was done next day after the first seedlings had emerged)



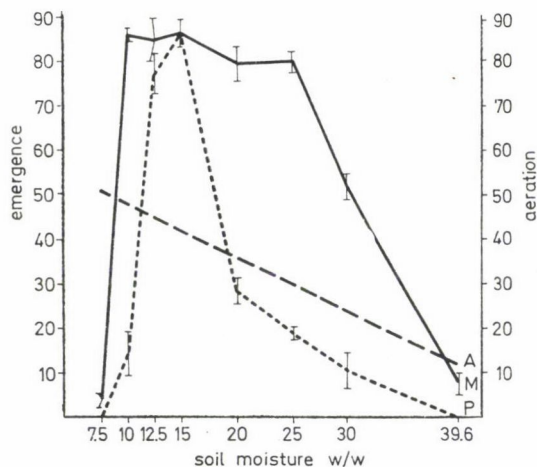


Fig. 2. Effect of soil moisture and aeration on the final emergence of raw and pelleted sugar beet seed. M ——— raw seed var. Monohybrid; P - - - pelleted seed; A = aeration

The effect of soil moisture on the seed's water uptake was studied in the second trial, which identically followed the method of the first one. After 24 hours, the seed was removed and its water content checked by weight. The moisture content of clusters and pelleting material was much higher than that of the soil: at soil moisture of 15%, the water content of pelleting material and clusters was 50.2% and 39.2%, respectively. At 5% soil moisture, it was 39.3% and 26.1%, respectively. The water uptake in the raw seed and in the clusters of pelleted seed was the same (39.4–39.2%) at soil moisture of 15%. At lower soil moisture, the water uptake in clusters of pelleted seed decreased more than in raw seed; at moisture of 5% it was 30.8% in the case of raw seed clusters and 26.1% of pelleted seed. This agrees well with lower emergence rates of pelleted seed at low soil moisture.

On the basis of our results and literary data, we were unable to assess whether the adverse effect of a high soil moisture is due to the inhibition of germination only in its very beginning or if the inhibition occurs at emergence or even later. To reveal the sensitivity of germs we arranged the third trial almost in the same way as the first one, except that instead of clusters, pregerminated seeds were planted (germs 5–10 mm long). Results presented in Fig. 3 demonstrate that the further growth of germs is not inhibited by a high soil moisture. Seed is very sensitive to high moisture at the beginning of germination. As soon as the seed has germinated, the germ is able to tolerate a higher moisture and lower aeration of the soil. Under such conditions, plants from pregerminated seeds emerged very quickly. A lower rate of germination in soil moisture under 25% was due to the higher mechanical resistance of the soil. Emergence at low soil moisture improved during the ensuing days.

Results presented in Figs 1 and 2 confirmed those schema of possible effects of various factors on germination that we have recently presented (VEVERKA 1982). Emergence follows the same trends as did germination at the various moisture regimes. Most important is the conclusion that our pelleted seed is able to emerge within a very narrow range of soil moisture content; in our trial it was only slightly more than from 12.5% to 15%. In comparison, raw seed performed well in a soil moisture range of 10–25%.

In laboratory tests presented recently (VEVERKA 1982) we came to the conclusion that "pelleting narrows down the extent of moisture conditions under which the seed is able



to germinate". This difference between raw and pelleted seeds is even more marked under soil conditions than in germination tests. The curve of emergence of pelleted seed is even sharper (Fig. 2).

An aeration as low as 31% was still nearly optimal for raw seed, emergence only decreased at a lower aeration. The optimum emergence of pelleted seed was reached at an aeration of 41%, and lower aeration emergence decreased very markedly. At an aeration of 31%, still suitable for the raw seed, the final emergence of pelleted seed was 19.0% only. Contrary to the raw seed, the emergence of pelleted seed was inhibited by an insufficient supply

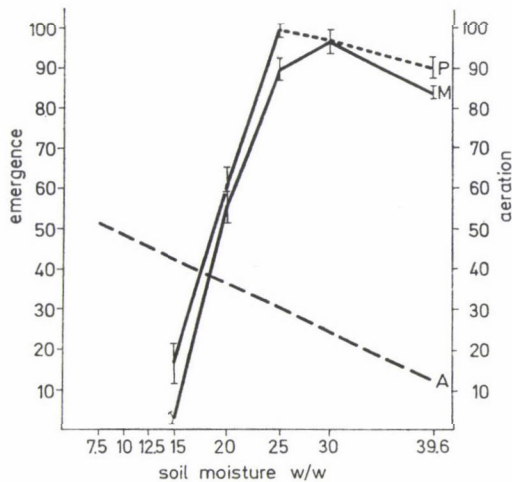


Fig. 3. Effect of soil moisture and aeration on the rate of emergence of pregerminated raw and pelleted sugar beet seed. M ——— raw, seed var. Monohybrid; P ——— pelleted seed; A = aeration. (Pregerminated seed with germ 1–10 mm long was planted: evaluation of the third day after planting)

of oxygen at soil moisture content higher than 12.5%. It was only at this moisture content higher than 12.5%. It was only at this moisture that no difference was recorded between raw and pelleted seeds (Fig. 2). However, the emergence rate was lower than at higher soil moisture (Fig. 1). This indicates that the 12.5% soil moisture content was below optimum and the seed did not receive sufficient water. Competitive relationship between water and oxygen uptake is even more important in soil conditions than in the germination tests.

Pelleted seed is reputed to be more susceptible to drought conditions than the original raw seed (HIBBERT, THOMSON and WOODWARK 1972). It also germinates less well in dry seed beds than does raw seed. We did not find any great difference in this respect from laboratory tests on blotting paper (VEVERKA 1982). In the present trials we have found very great differences in emergence, but only within a very narrow range of various moistures (Figs 1, 2). Pelleted seed reached almost the same emergence at soil moisture of 12.5% as did the raw seed at 10% moisture content. This accords well with the water content of the seed. At 15% soil moisture, the water content of clusters, after removal of pelleting material, was the same as that of raw seed. With decreasing soil moisture, the water content of clusters in pelleted seed decreased more than in raw seed (Table 1). These differences seem too small to

Table 1

*Effect of soil moisture on water uptake by raw and pelleted sugar beet seed*

Soil moisture W/W	15%		10%		5%	
Water uptake in	pelletting material	clusters	pelletting material	clusters	pelletting material	clusters
Raw seed		39.4		34.6		30.8
Pelleted seed	50.2	39.2	40.5	31.0	39.3	26.1

Water uptake is expressed in percentage of the weight of absolutely dry pelleting material or clusters

be of practical importance, but they agree with field experience. Under low-moisture regimes, very small change in water content causes great difference in water availability. Despite that, we assume greater importance to the sensitivity of pelleted seed to water surplus.

We studied both factors, water and aeration, under constant conditions only. Because field conditions vary, the fact of the intensivity of germs to water surplus is very important. The seed needs a relatively short time of suitable conditions to germinate and the ensuing growth can also proceed under high soil moisture. Such conditions are very dangerous because of fungal disease but in this paper we deal only with the simple ecological factors.

Our trials were done with one seed sample pelleted in one material by rolling process. Because they are in a good agreement with the previous results of laboratory tests reached also with other seed samples, different pelleting materials and foreign pelleted seeds (VEVERKA 1979, 1982), we can assume that our conclusions to some extent apply to every pelleting material and seed.

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#### References

- AURA, E. (1975): Effects of soil moisture on the germination and emergence of sugar beet (*Beta vulgaris* L.). J. Scient. Agric. Soc. Finl., **47**, 1–70.
- BLACK, C. A. (1968): Soil — plant relationships. John Wiley & Sons, Inc. New York—London—Sydney, p. 792.
- CURRIE, J. A. (1961): Gaseous diffusion in the aeration of aggregated soils. Soil. Sci., **92**, 40–45.
- FISCHNICH, O., GRIMM, H. and THIELBEIN, M. (1959): Einige pflanzen- und ackerbauliche Voraussetzungen für den Feldaufgang von Zuckerrüben — Monogermersaatgut. Zeitsch. f. Acker- und Pflanzenbau, Bd. **108**, 114–136.
- GARDNER, W. H. (1968): How water moves in the soil. Crops Soils, **21**, 7–12.
- HIBBERT, D., THOMSON, D. C. G. and WOODWARK, W. (1972): Some observation on the effect of different pelleting processes on the laboratory germination and field emergence of sugar beet seed. IIRB 35th Winter Congress, p. 135–144.
- HEYDECKER, W. and GULLIVER, R. L. (1972): Critical moisture conditions for the establishment of beet seedlings. 35th IIRB Winter Congress, p. 122–134.
- MEGARTHY, T. W. (1978): The physiology of seed hydration and the relation between water stress and the control of germination: a review. Pl. Cell Environ, **1**, 101–119.
- HUNTER, J. R. and ERICKSON, A. E. (1952): Relation of seed germination to soil moisture tension. Agron. J., **44**, 107–109.
- LONGDEN, P. C. (1973) after: GULLIVER, R. L. and HEYDECKER, W.: Establishment of seedlings in a changeable environment. In: HEYDECKER, W.: Seed ecology. London—Butterworths, p. 448.



- PERRY, D. A. (1973): Interacting effects of seed vigour and environment on seedling establishment. In: HEYDECKER, W.: Seed ecology, p. 311–323, London—Butterworths.
- ROSS, H. A. and HEGARTHY, T. W. (1979): Sensitivity of seed germination and seedling radicle growth to moisture stress in some vegetable crop species. *Ann. Bot.*, **43**, 241–243.
- STEHLÍK, V. and NEUWIRTH, F. (1928): Ökologie der aufgehenden Zuckerrübe mit besonderer Berücksichtigung ihrer Krankheiten. *Z. Zuckerind. Cechoslov. Republik*, **53**, 429–453.
- VANBREMEERSCH, P. (1972): Une nouvelle présentation des graines pour semoirs de précision. Le "pralinage" des graines de betteraves. *Proc. 35th Winter Congress IIRB*, Report No 20., Brussels.
- VANSTALLEN, R. (1971): L'influence de l'humidité sur la germination des graines enrobées. *Inst. Belge pour l'Amélioration de la Betterave* No. 4, 97–115.
- STOUT, B. A., SNYDER, W. and CARLETON, W. M. (1959): The effect of soil moisture and compaction on sugar beet emergence. *Journal of the Amer. Soc. Sug. Beet Technol.*, Vol. 9, 277–283.
- SNYDER, F. W. and ZIELKE, R. C. (1973): Water requirement for maximum germination and emergence of sugar beet seeds. *Journal of the Amer. Soc. Sug. Beet Technol.*, Vol. 17, 323–331.
- SPERLINGSON, CH. (1981): The influence of seed bed soil physical environment on seedling growth and establishment. *IIRB*, 44th Winter Congress, p. 59–77.
- VEVERKA, K., BREJCHA, V., LÖBL, F. and ZAHRADNÍK, K. (1979): Klíčivost obalovaného osiva cukrovky a ekologické faktory. *Agrochémia*, **19**, 209–213.
- VEVERKA, K. (1982): Effect of pelleting on water uptake and the germination of sugar beet seed. *Acta Agron. Acad. Sci. Hung.*
- WILLEY, L. A. (1970): Trials of commercial varieties of sugar beet. *Brit. Sugar Beet Rev.*, **10**, 165–170.
- WENGEL, R. W. (1966): Emergence of corn in relation to soil diffusion rates. *Agron. J.*, **58**, 69–72.

#### EFFECT OF IRRIGATION AND ANTITRANSPIRANTS ON NUTRIENT CONCENTRATION AND UPTAKE IN BARLEY

The concentration and uptake of nutrients in barley are influenced by a variety of factors, of which the moisture environment in root zone soil is important. SHARMA and SINGH (1973) observed a consistent increase in the nitrogen and phosphorus uptake due to increases in the soil moisture status of soil profiles. GRUNES (1959) also reported increased nitrogen and phosphorus uptake at low soil moisture stream in barley. However, the information on the effect of antitranspirants on nutrient uptake is inadequate, and the authors are aware of only one report in the literature about the influence of antitranspirant on nutrient uptake in barley (AGARWAL and DE 1979). This paper intends, therefore, to discuss the effect of different irrigation levels and use of various antitranspirants on the concentration and the uptake of nitrogen and phosphorus in barley.

A field experiment on barley (Cv. BG 25) was conducted during the winters of 1977–78 and 1978–79 at the Research Farm of Haryana Agricultural University, Hissar (India). The soil was sandy loam in texture, slightly alkaline in reaction (pH 7.8) and medium in fertility status (275 kg available N, 30 kg available  $P_2O_5$ , 360 kg available  $K_2O$ /ha). It retained about 24.5 cm and 10.5 cm water in one metre profile at 0.1 bar and 15 bar tensions, respectively. The bulk density of soil was  $1.44 \text{ g cm}^{-3}$ . The crop was sown after a uniform presowing irrigation of about 8 cm depth at 23 cm row spacing using 85 kg seed per hectare on 31st October for the 1977–78 season and on 14th November for 1978–79. The crop was harvested on 31st March, 1978 and 1st April, 1979 during the first and second season, respectively. The other agronomic practices and plant protection measures were followed according to recommendations.

The experiment was laid out in a split plot design with four replications. There were 24 treatment combinations comprising four irrigation levels in main plots ( $I_0$  — No post sowing irrigation;  $I_1$  — one irrigation at tillering;  $I_2$  — one irrigation at boot stage;  $I_3$  — two



irrigations; i.e. one at tillering and second at boot stage) along with six antitranspirant treatments in sub-plots ( $A_0$  — No antitranspirant,  $A_1$  — Atrazine 200 ppm,  $A_2$  — PMA  $10^{-4}$  M,  $A_3$  — Kaolin 6%,  $A_4$  — Atrazine 200 ppm + Kaolin 6%,  $A_5$  — PMA  $10^{-4}$  M + Kaolin 6%). A measured quantity of 6 ha cm water was applied at each irrigation. The post-sowing irrigation was applied at a crop age of 30 days in  $I_1$ , 81 days in  $I_2$ , and 30 and 81 days in  $I_3$  treatment during the two crop seasons. The antitranspirants were sprayed at two stages of crop growth, i.e. first at flower primordial initiation (45 days of sowing) and second at boot stage of crop development (80 days of sowing) of barley.

At harvest, the uptake of nitrogen and phosphorus was computed from the concentration of these nutrients in grain and straw and their respective yields. The concentration of nitrogen and phosphorus were determined colorimetrically.

#### *Nutrient concentrations*

**Nitrogen:** The nitrogen concentration in grain and straw did not differ markedly during these two seasons (Table 1). However, application of irrigation brought changes in the nutritional status of barley plant. An increase in the soil moisture supply due to frequent irrigations ( $I_3$ ) had minimum, and unirrigated barley maximum nitrogen concentration in grain and straw.

Table 1  
*Effect of irrigation and antitranspirants on nitrogen and phosphorus concentration in barley*

Treatments	Nitrogen concentration (%)				Phosphorus concentration (%)			
	Grain		Straw		Grain		Straw	
	1977-78	1978-79	1977-78	1978-79	1977-78	1978-79	1977-78	1978-79
<i>Irrigation levels</i>								
$I_0$	1.60	1.50	0.20	0.20	0.183	0.230	0.035	0.042
$I_1$	1.39	1.30	0.16	0.18	0.220	0.222	0.042	0.041
$I_2$	1.50	1.46	0.18	0.18	0.184	0.227	0.034	0.044
$I_3$	1.29	1.33	0.14	0.16	0.222	0.218	0.044	0.042
S.D.5%	0.10	0.09	0.02	0.01	0.025	N.S.	0.005	N.S.
<i>Antitranspirants</i>								
$A_0$	1.33	1.43	0.16	0.18	0.205	0.228	0.038	0.042
$A_1$	1.35	1.42	0.17	0.18	0.206	0.228	0.039	0.043
$A_2$	1.37	1.43	0.17	0.19	0.203	0.227	0.039	0.043
$A_3$	1.32	1.42	0.17	0.18	0.207	0.228	0.038	0.043
$A_4$	1.38	1.42	0.17	0.19	0.208	0.226	0.039	0.042
$A_5$	1.36	1.42	0.16	0.19	0.208	0.226	0.039	0.042
S.D.5%	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

Grain and straw also had significantly lower nitrogen concentration due to application of one irrigation at tillering ( $I_1$ ) than the application of single irrigation at boot stage ( $I_2$ ) or barley without any post-sowing irrigation ( $I_0$ ). The reduction in nitrogen concentration in barley under higher soil moisture status would have been merely a dilution effect due to better growth and yield of barley as reported by other workers (SHARMA and SINGH 1973, VERMA *et al.* 1976).

The use of antitranspirants did not affect the nitrogen concentration in grain and straw during two seasons significantly because the use of different antitranspirants did not have any bearing on growth (YADAV 1979) and productivity (Table 4) of barley.

**Phosphorus:** The concentration of phosphorus in barley plant increased due to an increase in soil moisture supply, i.e. being highest in  $I_3$  and lowest in  $I_0$  treatment (Table 1). However, the effects of irrigation on phosphorus content in grain and straw were negligible during 1978–79, which was wetter than the 1977–78 season. Higher moisture content in the root zone soil due to increased frequency of irrigation might have enhanced the mineralization and concentration of phosphorus in soil solution and thereby its greater uptake and concentration in grain and straw of barley. These results are in agreement with the findings of SHARMA and SINGH (1973).

The use of antitranspirants did not affect the phosphorus concentration significantly during any season.

### Nutrient uptake

**Nitrogen:** Nitrogen uptake in grain and straw was not affected significantly due to variation in irrigation treatments in 1977–78, because higher concentrations of nitrogen recorded in grain and straw of barley under  $I_0$  and  $I_2$  treatments were compensated by low yields under these treatments (Table 2). However, during 1978–79 the treatments with one irrigation at tillering ( $I_1$ ) and two irrigations at tillering and boot stage ( $I_3$ ) were statistically at par, but showed significantly higher nitrogen uptake as compared to the treatments without post-sowing irrigation ( $I_0$ ) and one irrigation at boot stage ( $I_2$ ). This was due to higher yields obtained in  $I_1$  and  $I_3$  treatments than in  $I_0$  and  $I_2$  treatments. These results are in agreement with the findings of SHARMA and SINGH (1973), VERMA *et al.* (1976) and BAJPAI

**Table 2**  
*Effect of irrigation and antitranspirants on grain and straw yield of barley*

Treatments	Grain yield (kg/ha)		Straw yield (kg/ha)	
	1977–78	1978–79	1977–78	1978–79
<i>Irrigation levels</i>				
$I_0$	2924	2727	5179	5181
$I_1$	3554	3491	6810	6960
$I_2$	3234	2634	5521	5125
$I_3$	3873	3176	7392	6677
S.D.5%	297	304	889	601
<i>Antitranspirants</i>				
$A_0$	3248	2961	6231	5906
$A_1$	3347	3022	6080	5988
$A_2$	3339	2958	6328	5878
$A_3$	3397	2978	6343	5972
$A_4$	3486	3027	6091	6033
$A_5$	3492	3095	6281	6137
S.D.5%	230	N.S.	N.S.	N.S.

and MERTIA (1977). The N uptake during two seasons was not affected significantly due to the use of various antitranspirants. AGARWAL and DE (1979) also reported insignificant effect due to PMA application on nitrogen uptake in barley.

The nitrogen uptake was significantly higher in  $I_1$  and  $I_3$  treatments as compared to  $I_0$  and  $I_2$  treatments during two seasons (Table 2). The minimum and maximum values of total nitrogen uptake varied between 56.64 and 61.98, and 48.43 and 58.42 kg/ha during 1977-78 and 1978-79, respectively in different treatments. The different antitranspirants did not vary markedly in respect to total nitrogen uptake and it ranged from 54.49 to 57.77 kg/ha during 1977-78, and 53.29 to 54.15 kg/ha during the 1978-79 season. The amount of nitrogen translocated to grain was more than 75% of the total uptake during the two seasons. The mean value was 82% and 78% during 1977-78 and 1978-79, respectively. The different irrigation and antitranspirant treatments hardly influenced the nitrogen translocation in grain by more than 2%. SINGH and SHARMA (1972) have reported up to 66% translocation of absorbed nitrogen by wheat plant to grain in the case of tall wheats.

**Phosphorus:** The uptake of phosphorus was affected significantly due to variation in irrigation treatments (Table 3). Application of one ( $I_1$ ) and two irrigations ( $I_3$ ) increased phosphorus uptake significantly over treatment receiving no irrigation ( $I_0$ ) and with one irrigation at boot stage of crop development ( $I_2$ ) during the two seasons. However, the differences between the former ( $I_1$ ,  $I_3$ ) and latter ( $I_0$ ,  $I_2$ ) set of treatments did not vary significantly. The increase in phosphorus uptake in grain and straw in  $I_1$  and  $I_3$  treatments was due to the additive effect of higher phosphorus concentration obtained in grain and straw and their higher yields than in  $I_0$  and  $I_2$  treatments. These findings are in general agreement with those of BAJPAI and MERTIA (1977) and SHARMA and SINGH (1973).

The treatment  $I_1$  and  $I_3$  showed significantly higher total phosphorus uptake as compared to  $I_0$  and  $I_2$  treatments during the two seasons. However, the differences between the

Table 3

*Effect of levels of irrigation and antitranspirants on nitrogen uptake (kg/ha) in barley*

Treatments	Grain		Straw		Total		N translocation to grain (%)	
	1977-78	1978-79	1977-78	1978-79	1977-78	1978-79	1977-78	1978-79
<i>Irrigation levels</i>								
$I_0$	46.23	40.55	10.41	10.35	56.64	50.90	81.62	79.67
$I_1$	51.92	45.35	11.16	13.07	63.08	58.42	82.15	77.63
$I_2$	47.78	38.46	9.62	9.97	57.40	48.43	82.81	79.41
$I_3$	51.26	42.42	10.33	12.16	61.59	54.58	83.64	77.72
S.D.5%	N.S.	4.11	N.S.	1.79	4.13	5.63		
<i>Antitranspirants</i>								
$A_0$	44.36	42.49	10.13	11.22	54.49	53.71	81.06	79.11
$A_1$	45.53	42.54	10.05	11.48	55.58	54.02	81.92	78.75
$A_2$	45.87	41.78	10.52	11.51	56.39	53.29	81.34	78.40
$A_3$	45.03	42.49	10.11	11.37	55.14	53.86	81.65	78.89
$A_4$	47.93	42.71	9.84	11.31	57.77	54.02	82.97	79.06
$A_5$	47.83	42.67	9.87	11.48	57.70	54.15	82.90	78.80
S.D.5%	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.		



Table 4

*Effect of irrigation and antitranspirants on phosphorus uptake (kg/ha) in barley*

Treatments	Grain		Straw		Total		P translocation to grain (%)	
	1977-78	1978-79	1977-78	1978-79	1977-78	1978-79	1977-78	1978-79
<i>Irrigation levels</i>								
I <sub>0</sub>	5.38	6.16	1.84	2.22	7.22	8.38	74.51	73.58
I <sub>1</sub>	7.86	7.84	2.83	2.93	10.69	10.77	73.52	72.79
I <sub>2</sub>	6.01	6.02	1.88	2.19	7.89	8.21	75.17	73.33
I <sub>3</sub>	8.67	7.43	3.29	2.84	11.96	10.27	72.49	72.35
S.D. <sub>5%</sub>	1.31	1.12	0.51	0.42	1.38	1.28		
<i>Antitranspirants</i>								
A <sub>0</sub>	6.92	6.75	2.43	2.48	9.35	9.23	74.01	73.13
A <sub>1</sub>	7.03	6.90	2.37	2.59	9.40	9.49	74.78	72.79
A <sub>2</sub>	7.06	7.01	2.48	2.50	9.54	9.51	74.00	73.71
A <sub>3</sub>	6.95	6.83	2.41	2.58	9.36	9.41	74.25	72.58
A <sub>4</sub>	7.01	6.81	2.38	2.55	9.39	9.36	74.65	72.76
A <sub>5</sub>	6.99	7.00	2.47	2.58	9.46	9.58	73.89	73.86
S.D. <sub>5%</sub>	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.		

former (I<sub>1</sub>, I<sub>3</sub>) and latter (I<sub>0</sub>, I<sub>2</sub>) set of treatments were not significant. The total phosphorus uptake did not vary markedly due to the use of various antitranspirants during the two seasons. The values of total phosphorus uptake varied from 9.35 to 9.54, and 9.23 and 9.58 kg/ha during 1977-78 and 1978-79, respectively. The percentage translocation of phosphorus to grain was affected neither by different irrigation levels nor by use of various antitranspirants. In general, about 74% of total phosphorus absorbed by the plant was translocated to grain and the remaining 26% remained in straw. SINGH and SHARMA (1972) reported 5% to 58% translocation of phosphorus to grain in tall varieties of wheat.

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#### References

- AGARWAL, S. K. and DE, RAJAT (1979): Effect of rates of nitrogen, mulching and antitranspirants on nutrient uptake of barley varieties under rainfed conditions. *Indian J. Agron.*, **24**, 66-72.
- BAJPAI, M. H. and MERTIA, H. S. (1977): A note on the irrigation and fertility levels on the yield and nutrient uptake of dwarf barley cultivar RDB-1. *Ann. and Zone*, **16**, 153-156.
- GRUNES, D. L. (1959): Effect of nitrogen on the availability of soil fertilizer phosphorus to plants. *Adv. Agron.*, **11**, 369-393.
- SHARMA, H. C. and SINGH, R. M. (1973): Effect of soil moisture regime, phosphorus and nitrogen on N and P uptake by barley (*Hordeum vulgare* L.). *Indian J. Agric. Sci.*, **43**, 58-61.

- SINGH, D. P. and SHARMA, H. C. (1972): The effect of different doses of nitrogen, phosphorus and potash on the growth, yield and quality of wheat. *J. Trop. Agri. and vet. Sci.*, **10**, 315-325.
- VERMA, S. V., MALIK, S. K. and AGARWAL, M. C. (1976): Effect of nitrogen application under varying soil moisture regimes on the uptake of nutrients in barley. *Indian J. Agron.*, **21**, 318-320.
- YADAV, S. K. (1979): Effect of levels of irrigation and use of antitranspirants on growth, yield, nutrient uptake and water use efficiency of barley (*Hordeum vulgare* L.). Thesis submitted to Haryana Agricultural University, Hissar (India).

### THE APPLICABILITY OF SOME PARAMETERS IN COMPARATIVE EVALUATION OF PHENOTYPIC CHARACTERS OF MAIZE HYBRIDS

The development of inbred methods in maize breeding is based on applied genetics as well as on the use of recent achievements of breeding methodology. Nevertheless one of the most essential and most difficult parts maize breeding is the evaluation of inbred lines and the composition of new, good combinations.

There are three hybrid evaluation parameters used generally by breeders, respectively: the correlation coefficient (COLLINS 1916, KIESSELBACH 1922, RICHEY 1924, HAYES 1926, HAYES and JOHNSON 1939, BROWN 1953, CAMACHO 1962, GAMA and HALLAUER 1977, etc.), the coefficient of variation (STRINGFIELD 1950, JONES 1958, SHANK and ADAMS 1960, KOVÁCS *et al.* 1970, etc.), and the value of heterosis index (LENG 1954, KOVÁCS 1969, WALTON 1971, DHILLON and SINGH 1977, etc.). It seems reasonable to determine not only these three parameters but their relations as well for the evaluation of phenotypic characters of maize hybrids.

The hybrids used in our trials were combinations of two female testers and 10 male lines. The inbred lines belonged to the IWGO program, and were as follows:

Male lines: NR 1065 (Austria), K 10 (Polish), EA 2087 (Spanish), ZPL 2077, ZP 2044-8 (Jugoslavia), F 478, Fr 106 (French), A 662 (USA), SL 442, TVA 1004 (Czechoslovakia).

Female lines: W 117 HT (USA), MR 21 (Hungary).

The trials were completed in 1976 and 1977 at Martonvásár, in 4 replications, in a randomized block design with a 70 × 30 cm growing area in 2-row plots of 20 plants each.

In the trials the following groups of characters were studied:

- a) Number of days from planting to 50% flowering;
- b) Morphological characters;
- c) Ear characters;
- d) Kernel characters.

The characters were studied from the data of 10 individual plants of each plot in all replications.

All data were evaluated by biometrical methods — analysis of variance, calculation of correlation indexes ( $r$  values), coefficients of variation (CV values) and heterosis indexes

$$\frac{\bar{P} - \bar{F}}{\bar{P}} \quad \text{— with HP 9831 A computer in Martonvásár.}$$

The variances, average values, limits of variability, as well as coefficients of variation and heterosis indexes of 19 characters of the  $F_1$  hybrids and their parents are presented in Table 1.

The analysis of variance of the two experimental years proves that the variance of the parents and the  $F_1$  hybrids for all 19 characters is significant (without the dry material

percentage value of 1976). The level of probability was 1% for kernel length (in case of parents in 1977) and for dry material percentage (in case of  $F_1$  hybrids in 1976). All the other characters were highly significant at  $P = 0.1$  per cent level.

The table shows that the strongest heterosis effect was found in grain yield formation. There was a considerable heterosis effect in the case of ear length, number of kernels per row, thousand-grain weight, plant height, ear height and leaf area. There was a heterosis effect indicating earliness in the case of tasseling and silking date. According to our experimental material, the mean heterosis index of grain yield (two years average) was 172.7%, which means that the yielding ability of the 20 single crosses was 2.72 times that of the parental lines. Our experimental result agree with JENKINS (1929), DAVIS (1934), JOHNSON and HAYES (1936), GREEN (1948), LONNQUIST (1953), KOVÁCS (1970).

The heterosis indexes of some characters of  $F_1$  hybrids, compared to their parental lines, were as follows:

ear height	189%,
number of kernels per row	181%,
leaf area	158%,
plant height	151%,
ear length	145%,
thousand-grain weight	123%.

The lowest values were found in the case of the number of days from planting to flowering (highly negative), number of leaves, number of ears per plant, shelling and dry material percentages. It was found that the  $F_1$  hybrids are earlier than their parental inbred lines. These results agree with STRINGFIELD (1950), SENTZ *et al.* (1954), ROW (1963), DANIEL (1974).

Generally, in the evaluation of phenotypic characters of hybrids, the three parameters — the heterosis index, the correlation coefficient and the coefficient of variation — are used separately. To supplement the evaluation process, together with the calculation of the three parameter values, their relations were also studied.

According to the 19 characters, it was stated in both years that the variability of  $F_1$  hybrids was lower than that of the homozygous parents. In these trials in the case of parents the mean coefficient of variation for the 19 characters was 23.70, but it was 12.18 for  $F_1$  hybrids. Similar results were obtained by STRINGFIELD (1950), SENTZ *et al.* (1954), JONES (1958), SHANK and ADAMS (1960), KOVÁCS *et al.* (1970). The coefficient of variation of ear diameter and shelling percentage were similar to that of the above-mentioned authors.

In addition, according to our results, the number of days from planning to flowering and the number of leaves appeared invariable. But examining the phenotypic variability of the 19 characters, the grain yield, leaf area, plant height, thousand-grain weight, number of kernel rows, number of leaves above the top ear, number of kernels per row, ear length and the dry material percentage proved to be variable characters.

The correlation of yield and other characters of the  $F_1$  hybrids was cleared in our examination. It was stated that the correlation between grain yield and other characters such as ear length, number of kernels per row, ear diameter, number of ears per plant, plant height, ear height, and leaf area was positively significant. Due to the correlation calculations, the value of the coefficients in  $F_1$  hybrids varied between 0.374+ to 0.831+++ for character pairs, respectively. This result agrees with that of NILSSON-LEISSNER (1927), JENKINS (1929), DAVIS (1934), LENG (1954), GENTER and ALEXANDER (1962), KOVÁCS (1969), KOVÁCS *et al.* (1970).



**Table 1**  
*Characters of F<sub>1</sub> hybrids and their inbred*

Characters	Year	Parents (inbred lines)			
		Variance		Limits of variability	Average values
		Treatments	Error		
(1)	(2)	(3)*	(4)*	(5)	(6)
Grain yield, g/plant	1976	444.46***	16.00	20.18– 55.68	34.69
	1977	247.68***	13.14	47.00– 74.95	63.83
1. Flowering,* day					
Male flowering	1976	30.28***	1.01	69.50– 77.50	73.18
	1977	29.74***	1.39	66.25– 74.75	69.73
Female flowering	1976	20.02***	1.53	77.58– 84.25	79.89
	1977	35.43***	1.43	66.00– 75.75	71.72
Average	1976	18.37***	0.94	74.00– 80.75	76.50
	1977	28.37***	1.27	66.38– 74.13	70.73
2. Morphological characters					
Plant height, cm	1976	895.04***	28.27	64.38–106.65	88.87
	1977	770.25***	28.30	105.78–152.52	130.21
Ear height, cm	1976	117.13***	7.05	22.10– 38.30	30.49
	1977	147.64***	12.43	40.25– 59.60	47.50
Total leaves	1976	2.58***	0.09	13.48– 16.18	14.87
	1977	1.86***	0.22	14.40– 16.75	15.56
Leaves above main ear	1976	1.92***	0.05	3.70– 5.38	4.76
	1977	1.78***	0.04	4.28– 5.98	5.03
Leaf area, cm <sup>2</sup>	1976	1 014 342***	18 619	1473–3026	2320
	1977	1 298 091***	20 970	1825–3485	2877
3. Ear characters					
Number of ears per plant	1976	0.37***	0.02	0.55– 1.68	0.97
	1977	0.15***	0.007	0.98– 1.62	1.16
Ear length, mm	1976	1555.23***	55.51	85.63–136.23	105.87
	1977	678.49***	21.83	120.28–158.28	139.84
Ear diameter, mm	1976	21.80***	2.74	29.75– 37.33	33.57
	1977	15.01***	0.88	31.20– 38.60	35.92
Cob diameter, mm	1976	14.81***	1.96	17.40– 23.30	20.78
	1977	19.99***	0.60	18.25– 25.53	23.24
4. Kernel characters					
Number of kernels per row	1976	73.72***	2.01	10.03– 24.40	19.32
	1977	47.25***	3.57	19.58– 29.30	21.16
Number of kernel rows	1976	9.66***	0.64	9.60– 14.88	13.07
	1977	17.16***	0.34	8.68– 16.75	13.73
Kernel length, mm	1976	1.77***	0.21	7.52– 9.82	8.53
	1977	1.10***	0.27	8.10– 9.60	9.16
Thousand-grain weight	1976	4585.07***	678.85	135.31–248.56	205.93
	1977	7024.38***	126.90	141.70–287.40	214.40
Shelling percentage	1976	186.40***	9.18	61.55– 85.93	80.14
	1977	21.17***	4.24	74.43–82.95	79.04
Dry material percentage	1976	99.95NS	44.70	56.55– 68.03	62.22
	1977	149.08***	29.57	61.58– 77.83	69.88

(3)\* FG: 11; (4)\* FG: 33; (8)\* FG: 19; (9)\* FG: 57

\* Number of days from planting to 50% flowering.

\*\* Significant at 1 per cent level

\*\*\* Significant at 0.1 per cent level

NS Non-significant difference

lines (Martonvásár, 1976-77)

F <sub>1</sub> Hybrids						
CV values (7)	Variance		Limits of variability (10)	Average values (11)	CV values (12)	$\frac{\bar{F} - \bar{P}}{\bar{P}}$ (13)
	Treatments (8)*	Error (9)*				
60.77	468.78***	46.12	102.10-138.71	116.42	18.57	2.356
24.66	216.46***	46.04	124.10-149.98	134.19	10.97	1.102
7.52	9.46***	0.84	68.00- 73.50	69.37	4.40	-0.052
7.81	7.70***	0.46	62.25- 67.00	64.81	4.28	-0.070
5.60	6.47***	0.82	71.00- 76.00	73.32	2.54	-0.091
8.29	18.51***	0.61	64.50- 72.50	68.17	3.98	-0.049
5.61	6.31***	0.57	69.38- 73.13	71.29	2.51	-0.068
7.54	9.66***	0.41	64.38- 69.75	66.48	4.68	-0.060
33.66	347.26***	47.55	131.38-158.73	146.58	12.72	0.649
21.32	505.63***	11.49	160.30-200.18	179.81	22.49	0.381
35.48	118.46***	14.45	50.60- 69.80	58.51	18.59	0.919
25.58	393.38***	9.00	73.63-106.38	89.08	22.26	0.875
10.82	0.99***	0.10	14.23- 16.25	15.42	6.49	0.037
8.74	1.35***	0.11	14.48- 16.53	15.78	7.35	0.001
29.76	1.09***	0.04	4.15- 6.00	4.95	21.17	0.042
26.64	0.91***	0.06	4.40- 6.05	5.08	18.89	0.010
43.40	453.752***	30 993	3598-4637	4016	16.77	0.731
41.47	776.006***	27 001	3521-4886	4151	21.22	0.442
62.82	0.0103***	0.002	0.93- 1.18	1.022	9.88	0.057
33.62	0.013***	0.0045	1.03- 1.22	1.082	10.54	0.067
37.23	411.04***	64.39	149.60-188.00	167.97	12.07	0.587
18.74	507.41***	50.98	172.03-212.53	183.58	12.31	0.313
13.91	11.37***	0.79	36.55- 43.78	40.84	8.25	0.217
27.31	7.78***	0.68	38.30- 43.78	41.09	6.78	0.144
18.52	15.74***	1.01	19.68- 27.80	23.95	16.58	0.153
19.23	11.02***	0.36	21.65- 27.40	24.59	13.50	0.058
44.40	27.38***	3.48	27.98- 39.33	35.27	14.83	0.826
10.77	17.51***	1.31	34.93- 42.05	38.30	10.94	0.810
23.76	7.14***	0.19	12.35- 17.65	15.19	17.58	0.162
30.15	9.40***	0.28	11.23- 17.25	15.58	19.70	0.135
15.63	1.51***	0.18	9.05- 11.10	10.06	12.23	0.179
11.46	1.09***	0.28	8.65- 10.83	9.91	10.60	0.082
32.76	3135.11***	169.67	216.19-297.81	264.64	21.21	0.285
39.09	2406.28***	74.12	217.21-289.28	254.41	19.23	0.187
17.03	9.37***	3.23	82.08- 88.83	86.25	3.55	0.076
5.82	10.03***	2.27	81.60- 87.50	83.90	3.78	0.061
16.05	27.88**	8.68	57.40- 69.50	63.90	8.26	0.027
17.47	42.99***	3.13	51.62- 63.65	57.28	11.45	0.180

In the complex line evaluation system the basis of the breeding value of lines is  $tF_1$  single cross. From the presented results on the evaluation of phenotypic characters of the hybrid maize, the following questions may arise:

a) Do characters of high heterosis index values have high variability?

b) What is the relation between grain yield and those characters which have strong heterosis effects?

c) Is it possible to combine the three parameters — the heterosis index, the correlation coefficient and the coefficient of variation — in hybrid comparative evaluation?

The comparison of the three parameters of  $F_1$  hybrid characters is shown in Table 2.

**Table 2**

*Relationship between the heterosis index values, the coefficients of variation and the correlation coefficients between the grain yield and 18 characters of  $F_1$  hybrids (Martonvásár, 1976–1977)*

Characters	$\frac{\bar{F} - \bar{P}}{\bar{P}}$	CV %	Correlation coefficients
Grain yield	1.727	20.47	—
Ear height	0.897	47.11	0.813***
Number of kernels per row	0.816	15.19	0.374*
Leaf area	0.586	19.26	0.475***
Plant height	0.515	24.11	0.739***
Ear length	0.450	15.03	0.417**
Thousand-grain weight	0.236	20.47	0.078 <sup>NS</sup>
Ear diameter	0.178	7.64	0.323*
Number of kernel rows	0.148	18.65	0.325*
Kernel length	0.130	11.42	0.144 <sup>NS</sup>
Cob diameter	0.104	15.12	0.153 <sup>NS</sup>
Dry material	0.103	14.71	—0.820***
Shelling percentage	0.068	4.64	—0.308*
Number of days to flowering	—0.064	8.14	—0.409**
Number of ears per plant	0.060	11.41	0.637***
Leaves above main ear	0.026	19.92	—0.103 <sup>NS</sup>
Total leaves	0.019	7.25	0.237 <sup>NS</sup>

\* Significant at 5% level

\*\* Significant at 1% level

\*\*\* Significant at 0.1% level

<sup>NS</sup> Non-significant difference

The ear height, plant height, leaf area, ear length and the number of kernels per row are characters of strong heterosis effect, are highly variable and highly correlated with grain yield. It is proved by the significant correlation coefficient and the high (above 20) or medium (from 10 to 20) coefficient of variation, and further the high (above 0.450 in this study) heterosis index values. It deserves notice that, among the characters, grain yield had the strongest heterosis effect and the highest variability.

The number of leaves, number of days from planting to flowering, shelling percentage and ear diameter are stable characters and, at least only in low correlation with grain yield, do not have significant heterosis effects.



The thousand-grain mass, number of kernel rows, dry material percentage, number of leaves above the top ear and cob diameter proved to be exceptional characters. Having high coefficients of variation, they show low heterosis index values, and their correlation with grain yield is rarely significant.

According to the comparison of those three main parameters the following conclusions can be drawn: 1. Some of the phenotypic characters are demonstrably variable and have strong heterosis effects, significantly correlated with grain yield. Conversely, some other characters proved stable, without any strong heterosis effects, and their correlation with grain yield is either negative, insignificant or rarely significant. 2. Breeders may combine all three parameters — the heterosis index, the coefficient of variation and the correlation coefficient with grain yield — in the evaluation of phenotypic characters, especially in the evaluation of  $F_1$  hybrids for maize breeding.

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### References

- BROWN, W. L. (1953): A summary of maize breeding techniques. *Tropical Agriculture*, **30**, 86–96.
- CAMACHO, L. H. (1962): Quantitative genetic analysis of physical components of yield in corn. Ph.D. Thesis. North Carolina State college. Department of genetics.
- COLLINS, G. N. (1916): Correlated characters in maize breeding. *Jour. Agr. Research*, **12**, 435–453.
- DANIEL, L. (1974): Studies on the inheritance of the flowering time in maize (*Zea mays* L.). *Cereal Research Comm.*, **2**, 227–236.
- DAVIS, R. L. (1934): Maize crossing values in second generation lines. *Jour. Agr. Research*, **48**, 339–357.
- DHILLON, B. S. and SINGH, J. (1977): Combining ability and heterosis in diallel crosses of maize. *Theor. Appl. Genet.*, **49**, 117–122.
- GAMA, E. E. G. and HALLAUER, A. R. (1977): Relation between inbred and hybrid traits in maize. *Crop. Sci.*, **17**, 703–706.
- GENTER, C. F. and ALEXANDER, M. V. (1962): Comparative performance of  $S_1$  progenies and test-crosses of corn. *Crop. Sci.*, **2**, 516–519.
- GREEN, J. M. (1948): Inheritance of combining in maize hybrids. *Jour. Am. Soc. Agron.*, **40**, 58–63.
- HAYES, H. K. (1926): Present day problems of corn breeding. *Jour. Am. Soc. Agron.*, **18**, 344–363.
- HAYES, H. K. and JOHNSON, I. J. (1939): The breeding of improved selfed lines of corn. *Jour. Am. Soc. Agron.*, **31**, 710–724.
- JENKINS, M. T. (1929): Correlation studies with inbred and crossbred strains of maize. *Jour. Agr. Res.* Vol. 39, **9**, 677–721.
- JOHNSON, I. J. and HAYES, H. K. (1936): The combining ability of inbred lines of Golden Bantam sweet corn. *J. Am. Soc. Agron.*, **28**, 246–252.
- JONES, D. F. (1958): Heterosis and homeostasis in evolution and in applied genetics. *Amer. Nat.*, **92**, 321–328.
- KIESSELBACH, T. A. (1922): Corn investigation. *Ner. Agr. Exp. Sta. Res. Bul.* 20.
- KOVÁCS, I. (1969): Study on breeding value of inbred lines (Beltenyésztett törzsek nemesítési értékének vizsgálata). Ph.D. Thesis, Martonvásár.

- KOVÁCS, I. (1970): Variation of combining ability in populations of long time inbred lines. Some methodological achievements of the Hungarian hybrid maize breeding. Akadémiai Kiadó, Budapest, 127–146.
- KOVÁCS, I., O'SVÁTH, J. and KOVÁCS, K. (1970): Analysis of the major phenotypic yield components in single cross WF 9 Ms×N 6 with the path coefficient method. Some methodological achievements of the Hungarian hybrid maize breeding. Akadémiai Kiadó, Budapest, 237–256.
- LENG, E. R. (1954): Effects of heterosis on the major components of grain yield in corn. Agron. Jour., **46**, 502–506.
- LONNQUIST, J. H. (1953): Heterosis and yield of grain in maize. Agron. Jour., **45**, 539–542.
- NILSSON-LEISSNER, G. (1927): Relation of selfed strains of corn to  $F_1$  crosses between them. Jour. Am. Soc. Agron., **19**, 440–454.
- RICHEY, P. D. (1924): Effects of selection on the yield of a cross between varieties of corn. U.S. Dept. of Agr. Bull. No. 1209.
- ROW, P. R. (1963): Phenotypic stability for certain characters in a systematic series of maize genotypes grown in different environments. Ph.D. Thesis. University of Wisconsin, Madison.
- SENTZ, J. C., ROBINSON, H. F. and COMSTOCK, R. E. (1954): Relation between heterozygosis and performance in maize. Agron. Jour., **46**, 514–552.
- SHANK, D. B. and ADAMS, M. W. (1960): Environmental variability within inbred lines and single crosses of maize. Jour. of Genet., **57**, 119–126.
- STRINGFIELD, G. H. (1950): Heterozygosis and hybrid vigor in maize. Agron. Jour., **42**, 145–152.
- WALTON, P. D. (1971): Heterosis in Spring Wheat. Crop. Sci., **11**, 422–424.

EFFECTS OF PHYTOHORMONES, GROWTH REGULATORS  
AND ANTIMETABOLITES ON THE GROWTH OF MYCELIUM  
AND CHARACTERISTICS OF DEVELOPMENT  
IN *ALTERNARIA HELIANTHI* (HANSF.) TUBAKI ET NISHIHARA  
AND *SCLEROTINIA SCLEROTIORUM* (LIB.) DE BARY

The effect of biologically active substances used in increasing quantities on fungi in practice is but little known. Few data are available on the interaction between various bio-active compounds and pathogenic microfungi. For the sake of orientation, we are therefore planning to carry on screening for the major phytopathogenic fungi. The compounds used are phytohormones, hormone-like regulators, synthetic cytokinin-like compounds, fungistatic substances and well-known antimetabolites of different nature. Our aim is to find efficient but not phytotoxic substances that can be used in practice, too. On the other hand, our data give a clue to the biochemical nature of the microfungi examined.

*Alternaria helianthi* and *Sclerotinia sclerotiorum*, two important sunflower pathogens were the test organisms of experiments evaluated in this paper. The former, a representative of *Deuteromycetes* causes a new, gradually spreading disease on leaf and stem. *Sclerotinia sclerotiorum* (*Ascomycetes*) is a pathogen either tolerant or resistant to most fungicides used in practice, due to the sclerotia, its characteristic resting stage, and to its hardly-known biochemical properties (TURNER 1971, APELT and BOCHOW 1975, STEADMAN 1979, WILLETS and WONG 1980).

The substances examined and the method are described below. Discs 0.25 cm<sup>2</sup> in size excised from one-week old cultures of the two pathogens grown on potato-dextrose-agar (pH 5.7) provided the inoculum.

The liquid medium was Lilly—Barnett's culture fluid slightly modified by us: 2.0 g glycine, 10.0 g D-glucose, 1.0 g  $KH_2PO_4$ , 0.5 g  $MgSO_4 \cdot 7H_2O$ , 1.0 mg  $FeSO_4 \cdot 7H_2O$ , 1.0 mg  $ZnSO_4 \cdot 7H_2O$ , 0.3 mg  $MnSO_4 \cdot H_2O$ , 5.0 µg biotin, 100.0 µg thiamin completed with distilled water to 1000 ml (pH 5.7).

The concentration of the screened substances was uniformly 10 mg/l. Name, abbrevia-



tion or symbol, and origin (producer) of the compounds used are: gibberellic acid = GA<sub>3</sub> (Phylaxia),  $\beta$ -indole-acetic acid = IAA (Loba), kinetin (Fluka), 2,4-dichloro-phenoxy-acetic acid = 2,4-D; benzimidazole (Fluka), 6-methyl uracil and R-MU (Isotope Institute of the Hungarian Academy of Sciences), naphthyl-acetic acid (Chemapol, Prague), colchicin (Reanal), Nystatin (Chinoin), allopurinol (United Pharmaceutical and Nutriment Factory), D-mannit (Reanal), 1,6-bis( $\beta$ -chloro-ethylamine)-1,6-dideoxy-D-mannit = Degranol; 1,6-dibromo-1,6-dideoxy-D-mannit = Myelobromol; 1,2,5,6-tetra-methanesulphonyl-D-mannit = Zitostop (Chinoin), 6-methylpurine; 6-mercaptopurine; 5-fluoro-uracil; 5-aminouracil; 5-bromo-6-methyl-uracil (Sigma), Sz-marked compounds (Isotope Institute of the Hungarian Academy of Sciences): Sz-6, Sz-6r, Sz-11, Sz-28, Sz-32, Sz-53, Sz-55.

The disinfectants (formaldehyde, mercuric chloride, Neomagnol or Chlorogen = benzene-sulphonic chloroamide-sodium, Sterogenol = cetyl-pyridinium bromide, Phenomerborum = basic phenyl-mercury-metaborate, Thiomersalum or Merthiolat = sodium-o-ethyl-mercury-II-thio -benzoate) are all commercially available preparations, of a purity corresponding to the prescriptions of the VI. Magyar Gyógyszerkönyv (Hungarian Pharmacopoeia).

The substances were dissolved in the culture fluid before sterilization. The solutions were distributed in Erlenmeyer flasks of 100 ml, 50 ml in each, with 4 replications per treatment. As a control the original culture fluid was used.

Inoculation was carried out in a B1 type laminar box after autoclaving. The flasks were incubated in an LP-103 thermostat for two weeks at  $25 \pm 1^\circ\text{C}$ .

Evaluation was carried out by weighing the mycelium (air dry weight). During the incubation period, on two occasions the growth characters were also surveyed.

In studying the phytotoxicity, the sunflower variety "Iregi Szürke Csíkos" (produced at Iregszemcse) was used as test plant. After surface disinfection (0.01% solution of HgCl<sub>2</sub> for 5 minutes, then washing in sterile water three times) 100 achenes per treatment were soaked for 24 hours in 10 mg/l concentration solutions of the compounds. Soaking in sterile water served for control. The achenes were then placed in Petri dishes on wetted filter paper and incubated at  $25 \pm 1^\circ\text{C}$  for 1 week.

Evaluation was carried out on the basis of the number of germinating achenes and the length of the radicle, and expressed in percentage proportion to the control.

The most efficient substances were examined for phytotoxicity with sunflower (ISZCS) and soya (ISZ-10) plants at the stage of 3-4 foliage leaves, too. The removed shoots were placed in solutions of 10 mg/l concentration and kept at room temperature in diffuse light for one week. Evaluation was carried out on the basis of symptoms of wilting.

### *I. Development characteristics of Alternaria helianthi on the basis of growth characters*

#### *1. Effects of phytohormones and growth regulators*

In the group of compounds, GA<sub>3</sub> increased the vegetative mass of mycelium to a great extent (Table 1). Colonies, developing from several growth centres, indicated that this concentration of GA<sub>3</sub> did not influence the germination of conidia, either. The colonies were superficial, felty, greyish-white with a darker substrate, similar to pigmentation under natural conditions.

Out of the other compounds of the group, 6-methyluracil, R-MU, IAA and benzimidazole inhibited the growth to a greater or lesser extent. The colonies developing from the conidia were mostly submersed, jelly-like, or the growth only developed around the inoculum (under the influence of IAA); that is, the germination of conidia was blocked. Kinetin caused complete inhibition.



Table 1

*Effects of phytohormones and growth regulators on the growth of mycelium in Alternaria helianthi (Hansf.) Tubaki et Nishihara and Sclerotinia sclerotiorum (Lib.) de Bary*

Name or symbol of compound	Air dry weight of mycelium in g and in % of the control			
	<i>Alternaria helianthi</i>		<i>Sclerotinia sclerotiorum</i>	
	g	%	g	%
GA <sub>3</sub>	0.1233	198.5	0.2977	193.8
Kinetin	0.0	0.0	0.0715	46.5
IAA	0.0364	58.7	0.1961	127.9
NAA	0.0744	120.0	0.1748	113.7
2,4-D	0.0659	106.2	0.2172	142.0
6-methyluracil	0.0186	30.0	0.1314	85.5
R-MU	0.0100	16.0	0.1956	120.3
Benzimidazole	0.0110	17.0	0.1461	95.1
Sz-6	0.0885	142.7	0.2605	169.6
Sz-6r	0.0720	116.1	0.1940	126.3
Sz-11	0.0752	121.2	0.3731	242.9
Sz-28	0.0635	102.3	0.2206	143.6
Sz-32	0.0577	93.0	0.2917	189.9
Sz-53	0.0	0.0	0.0	0.0
Sz-55	0.0359	57.8	0.1610	104.9
Control	0.0620	100.0	0.1536	100.0

Out of the regulators, Sz-6, Sz-11 and Sz-6r, as well as NAA, stimulated the mycelium formation to a greater or lesser extent (16–42 per cent). Mediumsize grey, felty colonies developed from a number of growth centres; besides them, many submersed, jelly-like colonies were formed. Pigmentation ranged from yellowish- to bluish-grey.

The herbicide character 2,4-D slightly stimulated the mycelium formation, but prevented the germination of conidia from falling off the inoculum, producing thereby a single thick, felty, superficial colony around the inoculum.

Sz-28 and Sz-32 did not bring about any considerable change; while Sz-55 greatly inhibited the growth, resulting in a limited volume of thin, gauzy, surface growth. Sz-53 totally prevented the development of the fungus.

## 2. Effects of antimetabolites

Colchicin hardly increased the mass of mycelium; however, the growth characters showed as much difference compared to the control as the colonies formed a continuous coating on the surface of the culture fluid (Table 2).

In our experiments Nystatin, a polien antimycotic, used in human therapy, had a high stimulative effect on the formation of vegetative mycelium. Colonies with thick, felty, bluish substrates were formed almost grown together on the surface. A medium amount of submersed growth also developed.

The uracil derivatives (5-fluorouracil, 5-bromo-6-methyluracil) reduced the amount of the developed mycelium, possibly by preventing the germination of conidia; namely the development of the colonies started from essentially fewer centres.

Table 2

*Effects of antimetabolites on the growth of mycelium in Alternaria helianthi (Hansf.) Tubaki et Nishihara and Sclerotinia sclerotiorum (Lib.) de Bary*

Name or symbol of compound	Air dry weight of mycelium in g and in % of control			
	<i>Alternaria helianthi</i>		<i>Sclerotinia sclerotiorum</i>	
	g	%	g	%
Colchicin	0.0649	104.6	0.1674	108.9
Nystatin	0.1056	170.3	0.2283	148.6
D-mannit	0.0560	90.3	0.1650	107.4
Degranol	0.0432	69.6	0.2158	140.6
Myelobromol	0.0510	81.9	0.1602	104.3
Zitostop	0.0645	103.2	0.1074	69.9
Allopurinol	0.0070	11.2	0.1437	93.5
6-Mercaptopurine	0.0110	17.0	0.0846	55.0
6-Methylpurine	0.0	0.0	0.0	0.0
5-Fluorouracil	0.0346	55.8	0.0003	0.1
5-Aminouracil	0.0313	50.8	0.1602	104.3
5-Bromo-6-methyluracil	0.0417	67.2	0.1587	103.2
Formaldehyde	0.0432	69.6	0.0743	48.3
Chlorogen	0.0469	75.6	0.1450	94.4
Sterogenol	0.0	0.0	0.0637	41.6
HgCl <sub>2</sub>	0.0	0.0	0.0	0.0
Phenomerbor	0.0	0.0	0.0	0.0
Merthiolat	0.0	0.0	0.0	0.0
Control	0.0620	100.0	0.1536	100.0

The xanthine-oxidase inhibitor allopurinol, the purine skeleton antimetabolites, the 6-mercaptopurine and the 6-methylpurine proved to be strong inhibitors. The colonies generally were submersed, jelly-like, and hardly pigmented. With 6-methylpurine present, the fungus did not start developing at all.

D-mannit and its cytostatic derivatives (Zitostop, Myelobromol, Degranol) were either slightly inhibitory or ineffective. The developing colonies were partly felty, superficial, partly medium-size, submersed, with a pigmentation lighter than in the control.

The known disinfectants (Sterogenol, Phenomerbor, Merthiolat) completely prevented the life functions of the fungus — as expected. HgCl<sub>2</sub> did not block the initial growth of the inoculum, but the growth did not reach any measurable amount, the conidia falling off the inoculum died without developing germ tubes.

Formaldehyde and chlorogen at the given concentrations caused but a minor degree of inhibition.

## II. Development characteristics of *Sclerotinia sclerotiorum* on the basis of the growth character

### 1. Effects of phytohormones and growth regulators

The mycelium-increasing effect of GA<sub>3</sub> could be observed in the case of *Sclerotinia sclerotiorum*, as well. IAA and R-MU also had a stimulatory effect, though to a lesser extent, as opposed to their influence on *Alternaria helianthi*. Thick, cottony colonies were formed

around the inoculum, then the sclerotium fundaments and the — mostly scattered — large, mature, black sclerotia appeared. The 6-methyluracil and the benzimidazole had a slight, the kinetin a stronger inhibition; but they all had less influence on *Sclerotinia sclerotiorum* than on *Alternaria helianthi*.

Some of the cytokinin-like regulators showed a strong stimulation on mycelium formation; the vegetative production was increased in the greatest measure by Sz-11, and in a lesser degree by Sz-32, Sz-6 and Sz-28. NAA, Sz-6r and Sz-55 also caused some increase; the latter had the opposite effect on *Alternaria helianthi*.

The explicit stimulative effect of 2,4-D could be also observed in this case. Beside a strong aerial mycelium formation, a relatively few, different-sized sclerotia developed.

The Sz-compounds caused certain changes in the culture characters, in comparison to the control. Aerial mycelium formation was more abundant. Furthermore, the sclerotia were mostly found irregularly scattered, with variance in size. On the surface of the control culture fluid, the sclerotia were mostly arranged in one or two concentric circles, and their shape generally was spherical or longish-oval.

In response to treatment with Sz-32, haptera formation was observed at the edge of the culture, adhered to the wall of the flask. In connection with *Sclerotinia trifoliorum* Eriks, this phenomenon is known, though the opinions on it differ (WILLETS and WONG 1980).

Out of the compounds of the group, Sz-53 was the only total inhibitor almost causing the lysis of the inoculum.

## 2. Effects of antimetabolites

Nystatin stimulated the growth of this fungus, too. It started the formation of a thick, woolly aerial mycelium; and later, large, scattered sclerotia appeared.

Colchicin also showed a stimulative effect, though to a lesser extent; under its influence, haptera were formed here, too.

Allopurinol had but a slight inhibitory effect, while that of the purine skeleton compounds 6-mercaptopurine was stronger. The former compound induced a thin, gauzy surface growth of mycelium with a small number of sclerotia; the latter, on the other hand, while producing a woolly aerial mycelium, prevented the formation of sclerotia in the phase of incubation that we studied. The 6-methylpurine caused — just as did *Alternaria helianthi* — a complete inhibition, almost the lysis of the inoculum.

The D-mannit and, among its derivatives the Zitostop, proved slightly inhibitory, while the Degranol had a stimulative effect. The inhibitory or stimulative effects of the other compounds of the group (D-mannit, Myelobromol) were negligible.

The uracil derivatives were found to be almost ineffective, with the exception of 5-fluorouracil, the presence of which caused a selective inhibition, at least in comparison with *Alternaria helianthi*.

Out of the mercury-containing disinfectants, both the HgCl<sub>2</sub> and the Phenomerbor and Merthiolat stopped the development of the fungus, and even the inoculum decayed. The formaldehyde had a slight inhibitory effect, but the chlorogen was almost totally ineffective.

Sterogenol — which completely prevented the growth of *Alternaria helianthi* — was better tolerated by this fungus.

## 3. Phytotoxicity examinations

Effects of 10 mg/l concentration solutions of the substances tested in the examination series on the germination of sunflower and length of radicle were studied (Table 3). The known phytohormones and regulators (GA<sub>3</sub>, IAA, 2,4-D, NAA) showed the expected effects. Sz-6r



Table 3

*Phytotoxicity of phytohormones, growth regulators and antimetabolites on the basis of their effect on germination in the sunflower variety ISZCS*

Name or symbol of compound	Germination % of achene and length of radicle in cm			
	Germination %		Length of radicle	
	average of 100 achenes	% of the control	in cm (average)	% of the control
<b>Phytohormones</b>				
GA <sub>3</sub>	41	52	8.45	145
IAA	15	19	3.20	55
<b>Regulators</b>				
NAA	9	11	7.30	125
2,4-D	0	0	0	0
Sz-6	44	56	5.24	90
Sz-6r	42	54	7.55	130
Sz-11	30	38	3.43	59
Sz-28	47	60	5.49	94
Sz-32	44	56	5.19	89
Sz-53	74	95	4.16	71
Sz-55	28	36	4.11	71
<b>Antimetabolites</b>				
Colchicin	59	75	3.47	60
Nystatin	60	77	5.19	89
5-Fluorouracil	52	66	5.86	101
5-Aminouracil	47	60	4.95	85
Formaldehyde	65	83	3.42	59
HgCl <sub>2</sub>	61	78	4.03	69
Control (average of 800 achenes)	78	100	5.82	100

increased the length of the radicle. Sz-53, which proved a total fungicide for both fungi, showed a negligible toxicity.

Effects of substances most active in the in vitro experiments on sunflower and soya shoots with 3–4 leaves were also examined. The results are contained in Table 4. The 5-fluorouracil and the Sz-53 had no toxic effect on the shoots. The kinetin and most of the antimetabolites, as well as the Sterogenol and the Merthiolat, when applied at a concentration of 10 mg/l, appeared to be highly phytotoxic.

Our preliminary studies have contributed new data to our knowledge of the effects of some bioactive compounds (partly well-known substances already known for their effects other than the ones studied here, partly new, synthetic compounds) on pathogenic microfungi.

It is remarkable that the widely used gibberellic acid and the 2,4-di-chloro-phenoxy-acetic acid, a compound of herbicide action, equally stimulated the development (mycelium growth, sclerotium and conidium production, respectively) of both microfungi examined by us. This fact by itself makes it reasonable to take the interrelations into thorough consideration.

Table 4

*Phytotoxicity of several phytohormones, growth regulators and antimetabolites to removed sunflower (ISZCS) and soya (ISZ-10) shoots with 3-4 leaves*

Treatments	Symptoms after 1 week of incubation	
	in sunflower	in soya
Control	symptomless	symptomless
Kinetin	complete withering	complete withering
Sz-6	wilting	symptomless
Sz-6r	initial wilting	initial wilting
Sz-11	symptomless	wilting
Sz-53	symptomless	symptomless
Zitostop	strong wilting	initial wilting
6-Methyluracil	complete withering	initial wilting
5-Fluorouracil	symptomless	symptomless
6-Methylpurine	complete withering	complete withering
Sterogenol	complete withering	complete withering
HgCl <sub>2</sub>	wilting	strong wilting
Merthiolat	complete withering	complete withering

On the basis of the results we may even draw the conclusion that the pathogenic microfungi can use the molecules, known to be antimetabolites, as anabolites (carbon or nitrogen sources) if they have enzyme systems capable of decomposing these compounds. It can be supposed that some substances are so inactivated that — as indifferent compounds — they are incorporated into the fungus' own cells.

The kinetin and the synthetic cytokinin-like (POZSÁR 1979) compounds used (6-methyluracil, R-MU, benzimidazole), when applied at the given concentration of 10 mg/l, are more or less of fungistatic effect.

Among the antimetabolites that inhibit the nucleic acid synthesis, the 6-methyl purine excels in action. It can be supposed that the nucleic acid synthesis of the microfungi is particularly responsive to this compound. The effect of 5-fluorouracil is interesting, because of its selective fungicide character as to the two species examined.

The effect of the non-phytotoxic Sz-53 is highly remarkable since it is a fungicide for both microfungi.

The interpretation of the effects of the structurally different Sz-compounds is a task of the future. Their influence on the protein synthesis is probably related to their special cell membrane-bound nature and their effectivity on the proton transfer system.

\*

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## References

- APELT, G. and BOCHOW, H. (1975): Zur profilaktischen Bekämpfung von *Sclerotinia sclerotiorum* de Bary bei Hausgurken. Arch. Phytopathol. Pflanzenschutz, **14**, 2.
- POZSÁR, B. I. (1979): Biological activity of 6-methyl uracil (6-MU) in comparison with pesticide-type derivatives. Acta Agron. Hung., **28**, 170–184.
- STEADMAN, J. R. (1979): Control of plant diseases caused by *Sclerotinia* species. Phytopathology, **68**, 904–908.
- TURNER, W. B. (1971): Fungal metabolites. Academic Press, London—New York.
- WILLETS, H. J. and WONG, J. A.-L. (1980): The biology of *Sclerotinia sclerotiorum*, *S. trifoliorum* and *S. minor* with emphasis on specific nomenclature. The Botanical Review, **46**, 102–156.

COMPARISON OF FODDER GRAINS (BARLEY, OAT  
AND SORGHUM VARIETIES) FOR NUTRITIVE VALUE ON THE BASIS  
OF LABORATORY ANALYSES AND FEEDING TESTS

## II. Feeding tests

In our first paper, 4 barley, 5 oat and 8 grain sorghum varieties were compared for nutritive value on the basis of laboratory analyses of their components. Our present paper gives an account of the results of feed conversion (digestibility), protein utilization and production tests of the same varieties performed parallel with the quality examinations. The feeding tests were carried out at the Debrecen University of Agricultural Sciences, Department of Animal Husbandry, in 1977 with rats as monogastric and sheep as polygastric model animals, and in 1978 with rats only. The fodder grain samples examined in the experiment were obtained from the previous year's trial plots of the following varieties and selections of the Cereal Research Institute (code: B = barley, O = oat, S = sorghum):

## 1977

B-1: GK-59 winter barley  
B-2: GK-awnless winter barley  
—  
O-1: Szegedi (30) early spring oats\*  
O-2: Condor spring oats  
—  
—  
S-1: Hybar 456 grain sorghum  
S-2: Hybar 242 grain sorghum  
S-3: GK-Tisza grain sorghum  
—  
—

## 1978

B-1: GK-59 winter barley  
B-2: Mutant two-row winter barley  
B-3: Horpácsi two-row winter barley  
O-1: Szegedi (30) early spring oats\*  
O-2: Szegedi winter oats  
O-3: GK-2 spring oats  
O-4: GK-3 (awnless) spring oats  
S-1: Tiszagyöngye grain sorghum  
S-2: Szegedi 200 grain sorghum  
S-3: Szegedi 613 grain sorghum  
S-4: Napsugár grain sorghum  
S-5: Remény grain sorghum

Since only the GK-59 winter fodder barley (B-1) and the Szegedi early spring oats (O-1) were included both years in the experiment, the results — similarly to those of the composition analyses — are presented for the two years separately. The varieties are compared

\* The prospective spring oat variety Szegedi 30 was given preliminary state certification by the Variety Qualification Council in 1978 under the name "Szegedi early" spring oats, and will hereafter be referred to by this name.



with one another, and the species evaluated on the average of the varieties. The objective of the experiment was to obtain informative data on as many varieties within the three grain species as possible. This, and the expenses of the experiments, were the reasons for the tests not being performed on sheep, in 1978.

Methodical problems of the monodiet feeding experiments will not be discussed in this place. The methodology, the digestion coefficients, the calculation formulae and methods of protein utilization indices, as well as the metabolism of the individual nutritive elements in the different animal species, are equally found in the respective chapters of HEROLD's (1977) text-book, and in the Hungarian and foreign literature of the subject.

#### *Utilization of nutritive elements by rats*

The rat is a model of monogastric animals. Tests performed on rats — as for the digestibility of feed — can be adapted to swine or even poultry.

The rat feeding tests were carried out with young female rats of about the same age (6–8 weeks) and weight (123–152 g) derived from isogene inbreeding. The animals were placed in metabolism cages constructed for this special purpose in groups of 5 per grain fodder sample.

The experiment consisted of the following phases:

- a) accommodation phase (6–7 days),
- b) preparation phase (7–9 days),
- c) examination phase (7–9 days).

Feeding and weight increase, as well as the amounts of urine and excrement, were measured by the group. The excrement collected carefully every day was sprayed with concentrated formic acid, then hermetically sealed and refrigerated. In the bottles used for collecting the urine, identical quantities of concentrated formic acid were measured at the beginning of each day of examination for the purpose of preservation. The daily amounts of urine were also sealed hermetically and refrigerated until processing.

The daily ration of a group was 50 g of sorghum, 75 g of barley and 100 g of oats. The rations of oats and barley were raised in consideration of their lower energy contents, but the amounts left over were carefully gathered and measured before the next feeding. This gave opportunity to ensure the isocaloric nature of each ration.

The average amounts consumed per group during the experimental period — as established after measuring the feed refused by the rats — were 580 g of barleys, 436 g of oats, 409 g of sorghum. (Although it was a case of monodiet feeding, from the amount of feed consumed one may still conclude certain preferences for or aversions to feed. Also, the results are to some extent distorted by the fact that the awned oats and barley were practically husked by the rats and only the caryopses consumed, although the husks pass mostly undigested anyway through their intestinal canals. This too shows how much the monogastric animals are sensitive to fibre.)

Utilization of nutritive elements, which are the digestion coefficients, are seen in Tables 1 and 2. As for the digestibility of total organic matters contained in the three cereal species, the oat varieties proved the best, followed by the sorghums, while the barleys were found to be the poorest. More or less the same can be said about the utilization of protein and fat. The inorganic (ash) components of the oats were particularly well utilized by the rats. Utilization of N-free extractable matters was considered sufficient in the case of all three cereals, which is an important fact, as in the kernels of cereals this nutrient fraction is the main carrier of the nutritive value.

**Table 1**

*Utilization of nutritive elements by rats (digestion coefficients, %)  
(1977)*

Code	Protein	Fat	Fibre	N-free extr.	Ash	Organic matter	Average*
B-1	76.94	48.83	—	94.76	72.76	90.36	76.73
B-2	70.00	57.50	—	93.55	74.77	88.52	76.87
Average	73.47	53.17	—	94.16	73.77	89.44	76.80
O-1	86.86	88.46	—	73.51	96.00	92.30	87.43
O-2	82.44	77.57	—	69.90	96.31	91.13	83.47
Average	84.65	83.01	—	71.70	96.15	91.72	85.45
S-1	72.33	70.32	—	92.75	54.89	87.53	75.56
S-2	84.36	89.63	49.98	95.07	44.74	91.94	81.15
Average	78.35	79.98	—	93.91	49.82	89.74	78.36
Species average	78.82	72.05	—	86.59	73.25	90.30	80.20

\* Without the fibre content

**Table 2**

*Utilization of nutritive elements by rats (digestion coefficients, %)  
(1978)*

Code	Protein	Fat	Fibre	N-free extr.	Ash	Organic matter	Average*
B-1	71.5	48.8	—	90.9	55.7	83.5	70.1
B-2	72.0	50.9	—	94.0	40.2	87.3	68.9
B-3	75.2	44.3	4.5	93.2	62.0	86.9	72.3
Average	73.0	48.0	—	92.7	52.6	85.9	70.4
O-1	88.8	88.7	—	97.7	71.9	93.3	88.1
O-2	89.1	88.7	31.6	98.8	78.5	94.8	90.0
O-3	92.7	84.5	—	96.2	76.0	93.8	88.6
O-4	89.4	82.8	—	97.4	75.9	91.9	87.5
Average	90.0	86.2	—	97.5	75.6	93.5	88.6
S-1	67.6	86.1	44.3	92.5	51.2	87.8	77.0
S-2	67.5	86.3	46.0	92.1	30.0	87.6	72.7
S-3	88.7	88.2	68.4	97.8	69.5	95.8	88.0
S-4	69.7	87.3	36.6	93.3	65.1	89.7	81.0
S-5	73.6	84.7	31.2	90.8	43.5	89.9	75.9
Average	73.5	86.5	45.3	93.3	51.9	89.5	78.9
Species average	78.8	73.6	—	94.5	60.0	89.6	79.3

\* Without the fibre content

*Barleys, 1977*

Protein, N-free extractable material and total organic matters were best utilized by the GK-59, while the GK-awnless variety excelled in the digestibility of fat and ash content. Digestibility of fibre was not perceptible in both of the varieties examined.

1978

The utilization data of nutritive elements obtained by us for the three barley varieties largely agree with the values in the table for Hungarian standards. As regards the utilization of organic matter, the Mutant two-row barley was found to be the best, the Horpácsi two-row variety was similarly good, while the organic matter content of the GK-59 winter barley was somewhat less digestible. As for the digestibility of raw fibre, acceptable value was obtained only in the case of the Horpácsi two-row variety; it corresponds with the average value contained in the standard table.

*Oats, 1977*

The digestibility of all organic nutrients was the best in the variety Szegedi early. In the utilization of ash components there was no considerable difference between the varieties. The digestibility of fibre could not be determined again. The fibre consumed was practically fully excreted with the faeces.

1978

Utilization of the nutritive elements of oat samples in our experiment was found without exception to be better compared with the values of the Hungarian fodder table. The rats digested the protein and fat particularly well, the fibre very poorly and the N-free organic matter sufficiently well. As for the utilization of the total organic matter content, no great differences were observed. Yet, Szegedi winter oats proved the best and GK-3 spring oats the poorest. For Szegedi early and GK-2, medium values of organic matter utilization were obtained.

Further facts revealed by Tables 1 and 2: the oats consequently exceeded the species' average in both years, and their digestion coefficients showed the least variation.

*Sorghums, 1977*

(Since the sorghum sample GK-Tisza arrived late, it was not included in the experiment.) The digestibility of protein, fat, N-free extractable matter and total organic matter was the best in the case of the variety Hybar 242. Only in the utilization of the ash components was the hybrid sorghum Hybar 456 somewhat superior to it. The high tannin content of the latter supposedly by itself caused insufficient digestion. The digestion coefficient for the fibre content could only be established with Hybar 242.

1978

The utilization values for nutritive elements in the examined sorghum varieties generally are higher than those of the Hungarian fodder table. The variety Szegedi 613 proved the best in respect to all nutritive elements, though the data of "Napsugár" are also good. Neither were the other varieties found much inferior concerning the utilization of the total organic matter content. (There are improbably low values but they are partly regarded as experimental errors.)



*Protein utilization studies on rats*

The protein utilization studies were carried out parallel to the other utilization tests on the same experimental animals.

The utilization of protein was evaluated as a function of absorbed and excreted nutrients. (The quantity and composition of faeces and urine are not given here.)

While in 1977 the crude protein content of feed was best utilized by the rats in the case of oats and least with the barley varieties, in 1978 the average value was the highest for sorghum, and the lowest for barley again. The utilization of the absorbed (digestible) protein was the best in 1977 in the case of oats and the worst with sorghums. In 1978 the order of species was the same as in the case of crude protein utilization. It should be noted, however, that — for economic reasons — as regards the utilization of protein, the crude protein is more important than the digestible one. At the same time, on establishing the biological value of protein, the utilization of the digestible, more exactly of the absorbed protein, is taken for basis primarily (Tables 3 and 4).

**Table 3**  
*Protein utilization in rats ( $n = 5$  per group)*  
(1977)

Code	Protein taken up with feed	Protein excreted			Absorbed protein	Retained protein	Protein utilized as percentage of	
		with faeces	with urine	total			crude	digestible
		g					protein, %	
B-1	97.78	22.55	9.24	31.79	75.23	65.99	67.49	87.72
B-2	92.53	27.78	12.56	40.34	64.75	52.19	56.40	80.60
Average	95.16	25.17	10.90	36.07	69.99	59.09	61.95	84.16
O-1	99.50	13.07	12.75	25.82	86.43	73.68	74.05	85.25
O-2	86.18	15.13	9.43	24.56	71.05	61.62	71.50	86.73
Average	92.84	14.10	11.09	25.19	78.74	67.65	72.78	85.99
S-1	82.69	22.88	9.50	32.38	59.81	50.31	60.84	84.12
S-2	87.30	13.65	16.25	29.90	73.65	57.40	65.75	77.94
Average	85.00	18.27	12.88	31.15	66.73	53.86	63.30	81.03
Mean total	91.00	19.18	11.62	30.80	71.82	60.20	66.01	83.73

*Barleys, 1977*

Of the two barley varieties examined, it was GK-59 in which the protein content was far better utilized, whether the total crude or the absorbed protein was considered.

*1978*

As a somewhat contradictory result, the crude protein utilization of GK-59 was then found to be rather poor. The Mutant two-row winter barley was the best. The order of varieties was the same concerning the utilization of digestible protein.

**Table 4**  
*Protein utilization in rats (n = 5 per group)*  
 (1978)

Code	Protein taken up with feed	Protein excreted			Absorbed protein	Retained protein	Protein utilized as percentage of	
		with faeces	with urine	total			crude	digestible
		g					protein, %	
B-1	53.0	15.1	24.8	39.9	37.9	13.1	24.7	34.6
B-2	49.7	13.9	14.7	28.6	35.8	21.1	42.5	58.9
B-3	63.0	15.6	22.5	38.1	47.4	24.9	39.5	52.5
Average	55.2	14.9	20.7	35.5	40.4	19.7	35.6	48.7
O-1	95.8	10.7	31.9	42.6	85.1	53.2	55.5	62.5
O-2	92.9	10.1	38.1	48.2	82.8	44.7	48.1	54.0
O-3	106.2	7.7	27.6	35.3	98.5	70.9	66.8	71.9
O-4	119.5	12.7	34.2	46.9	106.8	72.6	60.8	60.7
Average	103.6	10.3	33.0	43.3	93.3	60.4	57.8	62.3
S-1	27.5	8.9	1.8	10.7	18.6	16.8	61.1	90.3
S-2	27.1	8.8	2.4	11.2	18.3	15.9	58.7	86.9
S-3	27.6	3.1	3.6	6.7	24.5	20.9	75.7	85.3
S-4	22.8	6.9	2.2	9.1	15.9	13.7	50.1	87.2
S-5	34.9	9.2	3.3	12.5	25.7	22.4	64.2	87.2
Average	28.0	7.4	2.7	10.0	20.6	17.9	62.0	87.2
Mean total	62.3	10.9	18.8	29.6	51.4	32.7	51.8	66.1

#### *Oats, 1977*

The protein of Szegedi early was better utilized than that of Condor. This was a remarkable result because the amino acid composition of the latter was found to be more favourable than that of the former.

1978

The total crude protein content was best utilized from GK-2, then from GK-3, less so in the case of Szegedi early and least of all with the Szegedi winter oats. As for the utilization of digestible protein, the order of varieties was different: GK-2 was invariably the first, followed by Szegedi early, then GK-3, and the Szegedi winter oat variety was the last again.

#### *Sorghums, 1977*

As regards the utilization of crude protein the variety Hybar 242, while in respect of absorption of protein the variety Hybar 456 proved to be better by 8–10%, due to its higher essential amino acid content.

1978

The five sorghum varieties were equally very good from the standpoint of total crude protein utilization; Szegedi 613 was best, "Remény" next best, and "Napsugár" relatively poorest. The utilization of digestible protein contained in them was also good. The best result was attained with "Tiszagyöngye", the least favourable with Szegedi 613. The high rate utilization of proteins of "Remény" could be foreseen because of their relatively favourable amino acid composition.

*Production studies, weight increase*

The groups of 5 animals were compared for weight increase by the kind of fodder grain fed. Table 5 shows the percentage weight increase in the examination phase only.

**Table 5**  
*Weight increase of young rats during the metabolism experiment*

Code	Average weight increase per animal during the period of examination, %		Two years' average*
	1977	1978	
B-1	9.2	21.2	15.2
B-2	10.5	13.9	
B-3	—	18.1	
Average	9.9	17.7	13.8
O-1	3.1	5.2	4.2
O-2	10.8	18.5	
O-3	—	—1.9	
O-4	—	8.5	
Average	7.0	7.6	7.3
S-1	1.7	2.3	
S-2	0	2.3	
S-3	—	2.2	
S-4	—	—6.7	
S-5	—	—7.0	
Average	0.9	—1.4	—0.3

\* Note: Since only the varieties B-1 and O-1 were included both years in the experiments the results of the other varieties cannot be averaged.

Although the figures of weight increase give most suggestive information about the actual production value of each cereal species, comparison is worth being made only among the varieties, since — as we have mentioned before — the experimental groups did not consume identical quantities from the three maize species. Isocalorism could, though, be ensured to some extent by equalizing the rations; yet, the unfavourable effect of monodietic feeding on



the appetite of the animals (gastro-intestinal depression, etc.) is very difficult — if possible at all — to eliminate (as indicated among others by the negative figures of weight increase). So it is only the varieties of the same corn species that can be rationally compared, although the order of species was the same in the two years and corresponded to the quantities of feed consumed (barleys: 580 g; oats: 436 g; sorghums: 409 g).

Of the barley varieties, the GK-59 winter barley resulted in the greatest weight increase, nor was the result of Horpácsi two-row much worse. A slightly lower weight increase was obtained with the Mutant two-row barley. In 1977 the GK-awnless gave a somewhat better result than the GK-59.

Of the oat varieties, the Szegedi winter oats resulted in the highest increase of weight. Condor was next best (in 1977) followed by Szegedi early. Weight decrease rather than increase was observed when the prospective variety GK-2 was fed.

Sorghum gave the poorest results. The weight reduction observed in the case of "Napsugár" and "Remény" was obviously the consequence of the previously mentioned bad effect of monodietic feeding on appetite. The relatively high tannin content of sorghum is also disadvantageous in feeding monogastric animals.

#### *Nutrient utilization in sheep*

Sheep feeding tests were carried out in metabolism pens, one for each animal. The experimental stock consisted of 10-week old merino lambs of about 20 kg in weight, 3 per each kind of fodder grain. The experimental period was divided into two phases; one week of preparation, and one week of examination. In the examination phase, we measured the average quantities of feed consumed. The amount of feed consumed per animal, as averaged after the remaining quantities were measured back, was 3163 g of barley, 3480 g of oats and 3689 g of sorghums. The results of utilization experiments, which are the digestion coefficients, are contained in Table 6.

**Table 6**  
*Utilization of nutritive elements by sheep (digestion coefficients, %)*  
(1977)

Code	Protein	Fat	Fibre	N-free extr.	Ash	Organic matter	Average*
B-1	86.86	81.07	—	90.18	9.39	86.19	70.74
B-2	83.12	86.95	—	91.41	36.54	86.98	77.00
Average	84.99	84.01	—	90.80	22.97	86.59	73.87
O-1	87.80	91.30	—	78.36	37.17	72.26	73.38
O-2	84.51	90.01	—	79.22	60.72	73.04	77.50
Average	86.16	90.66	—	78.79	48.95	72.65	75.44
S-1	84.28	92.94	18.59	83.37	77.52	82.75	84.17
S-2	85.19	95.75	77.47	93.26	73.69	91.77	87.93
S-3	69.90	94.35	84.77	84.81	76.84	83.82	81.94
Average	79.79	94.35	60.28	87.15	76.02	86.11	84.68
Species means	83.65	89.67	—	85.58	49.31	81.78	78.00

\* Without the fibre content

On the basis of the average values of digestion coefficients, we can say that as regards protein the highest rate of utilization was achieved with oats; while in the case of fat sorghums, and when the N-free extractable material and total organic matter are considered, barley varieties (the awnless one in particular) were best utilized. The extraction of fibre was only possible in the case of sorghums, so the digestion coefficients too are only given for sorghums; but the wide scatter of data show that they are of mere informative character.

Among the barley varieties GK-59 was the best concerning the digestibility of protein, while the utilization of the other nutritive elements was better in the case of GK awnless barley fed.

Among the oats, Szegedi early resulted in the best utilization of protein and fat, while the digestibility of N-free extractable material, ash and total organic matter content was more favourable with the variety Condor.

Among the sorghums, Hybar 242 was found to be the best and GK-Tisza the poorest in regard to the digestibility of nutrients; the high rate of utilization of fibre contained in these varieties is remarkable.

#### *Protein utilization in sheep*

The protein utilization tests were performed at the same time with the nutrient utilization experiments, parallel to them, on the same experimental animals. (The indices of protein utilization are contained in Table 7.)

**Table 7**  
*Protein utilization in sheep (n = 3 per group)*  
(1977)

Code	Protein taken up with feed	Protein excreted			Absorbed protein	Retained protein	Protein utilized as percentage of	
		with faeces	with urine	total			crude	digestible
		g					protein, %	
B-1	410.90	54.01	49.37	103.38	356.89	307.52	74.84	86.17
B-2	325.16	54.90	81.62	136.52	270.26	188.64	58.02	69.80
Average	368.03	54.45	65.50	119.95	313.58	248.08	66.43	77.98
O-1	458.85	56.25	15.76	72.01	402.60	386.84	84.31	96.09
O-2	398.75	61.21	12.74	73.95	337.54	324.80	81.46	96.23
Average	428.80	58.73	14.25	72.98	370.07	355.82	82.88	96.16
S-1	493.08	77.53	72.44	149.97	415.55	343.11	69.59	82.57
S-2	504.00	74.65	151.94	226.59	429.35	277.41	55.04	64.61
S-3	311.64	93.81	49.50	143.31	217.83	168.33	54.01	77.28
Average	436.24	82.00	91.29	173.29	354.24	262.95	59.55	74.82
Mean total	411.02	65.06	57.01	122.07	345.96	288.95	69.62	82.99

As mentioned with the rat feeding tests, the utilization of protein is measured as a function of absorbed and excreted nutrients; namely, the amount of urine, the N content discharged with urine, and the composition of faeces, are all of decisive importance. (Data on urine and faeces are not presented here, nor is the weight increase of sheep, the latter for deficiencies of data surveying.)

As for barley, a much better utilization of protein was obtained again with the GK-59, just as in the rat experiment. The same applies to the absorbed protein.

In the case of oats, the utilization of crude protein — as in the rat experiments — was more favourable with the Szegedi early; as regards the utilization of digestible protein no considerable differences were found among the varieties.

Of the sorghums in this case — in contrast to the rat experiments — the crude and, digestible protein contents of the variety Hybar 456 were better utilized. The utilization of digestible protein from the GK-Tisza can be said to be of medium rate.

To summarize, it can be established for all three varieties examined that the crude protein was better utilized by the sheep, the digestible protein more or less equally by rats and sheep, although the outstanding values obtained with oats confirm the important role played by this fodder grain species in the diet of ruminants.

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### References

HEROLD, I. (1977): Takarmányozás (Feeding). Mezőgazdasági Kiadó, Budapest.

### BEHAVIOUR OF COWS IN HEAT UNDER LARGE-SCALE CONDITIONS

It is known that in female animals the effect of the oestrogenic hormones on the genitalia manifests itself not only in the oestral symptoms but also in the behaviour of the animal.

In dairy farms the optimum time between calvings is very difficult to keep, though the prolongation of this interval causes considerable economic losses. A frequent cause of a longer than desired period between calvings is that the oestrus is unnoticed and so the insemination after calving does not take place in time.

According to the results of investigations in Canada (KING and HURNIK 1979), of the cows inseminated after having been marked with the so-called "marker" bull, 64 per cent conceived; while, of those detected by the tenders to be in heat, only 45 per cent became pregnant after the first insemination.

It is thus a highly important task to follow the time and intensity of the oestrus with attention, as this is the only way to avoid missing conception. With the increasing concentration of cattle stocks, on the other hand, it is more and more difficult to select the cows in heat either in bound- or in free-pen keeping.

By studying the behaviour of cows in heat, we wanted to discover the effect of industrial industry-like, keeping, and technological systems on the measureable, or at least perceptible, oestral symptoms; whether there was any difference in this respect between genotypes and keeping systems; and what were the correlations between the measurable oestral symptoms and the standard qualities.



The investigations were made in Hungarian red spotted, Holstein-Frisian and "Hungaro-Frisian" cattle stocks. The "Hungaro-Frisian" stock was studied in tied keeping while the other two breeds in tied and loose keeping alike. The observations covered the length of the oestral period (including the point of time when the vaginal orifice becomes swollen and when a thread of mucus appears and disappears in its corner), the intensity of heat, and the activity and behaviour of animals in heat.

In the course of the observations we found the period of perceptible oestrus to be shorter in the Hungarian red spotted and longer in the Holstein-Frisian and "Hungaro-Frisian" populations (Table 1). As to the length of the oestral period, differences between the

**Table 1**  
*Length of the oestral period and time of its appearance*

Designation	n	Length of the period of oestrus			Time of beginning of oestrus, %	
		x	±s	V%	at dawn	in the evening
<hr/>						
Hungarian red spotted						
bound system	56	14.4	9.1	63.19	76.2	15.8
free-pen system	42	13.1	9.5	72.5	74.1	13.3
Holstein-Frisian						
bound system	72	21.4	9.0	42.05	79.2	12.3
free-pen system	86	19.8	8.6	43.4	71.6	14.6
“Hungaro-Frisian”						
bound system	51	21.5	6.9	32.09	77.1	15.4

bound- and free-pen keeping systems were observed in the Hungarian spotted and Holstein-Frisian breeds. The oestrus mostly started in the early morning hours. The first symptoms in the bound keeping system were: during the night the cow repeatedly lay down for 20–30 minutes, then stood up and placed her head over the other cow's neck. One or two hours after these behaviour patterns, the vagina was seen to swell.

In the free-pen system the oestrus begins in most cases at the same time. Observation is more difficult here than in the bound keeping system. An indication of the beginning of the oestrus is that, while the other cows lie down at night, those on the verge of heat remain standing, then try to get into contact with their companions (by jumping over them, licking them and rubbing against them). Simultaneous with these behaviour patterns the swelling of the vagina can be observed (Fig. 1).

The fact that the symptoms of oestrus in the Hungarian red spotted breed last for a shorter time does not necessarily mean that the oestrous cycle of the breed is shorter than that of the Holstein-Frisian cattle. To decide this question, the oestrogenic levels should first be determined. It is in any case remarkable that, because of the shorter duration of visible symptoms of oestrus, animals in heat in this breed are more difficult to detect than in the Holstein-Frisian breed or Hungaro-Frisian construction.

The mass appearance of the beginning of oestrus in the early hours after midnight differs to some extent from the observations of HURNIK and KING (1979) who found that in free-pen keeping most cows showed the first signs of being in heat between 6 and 12 p.m. These differences may be supposed to arise from methodological differences.

In Fig. 1 the trend of intensity of oestrous symptoms in a bound system of keeping is seen. A very poor symptom of oestrus is spoken of when the vagina is but slightly swollen; the animal, though looking here and there, is quiet, and the rectal examination points out a small amount of oestrous mucus. The oestrus is intensive when the vagina is conspicuously swollen, an abundance of oestrous mucus appears mostly hanging out, the vaginal wall reddens, the animal keeps stamping about, raising its tail, and mooing. The poor and medium intensive symptoms are between these two extremes.

Data shown in Fig. 1 reveal that 17 per cent of the Hungarian red spotted cows and 8 per cent of each of the Holstein-Frisian and Hungaro-Frisian exhibit very poor oestrous symptoms. Intensive oestrous symptoms have been found in 28 per cent of the Hungarian

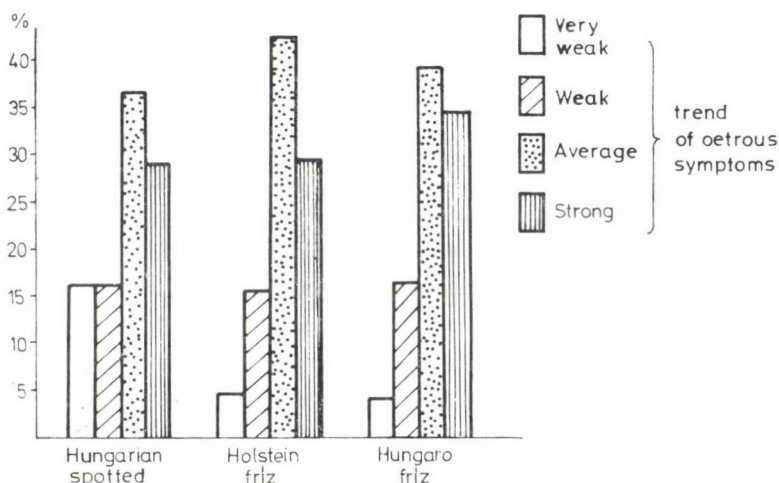


Fig. 1. Trend of intensity of oestrous symptoms in a bound keeping system

spotted cows, 29 per cent of the Holstein-Frisian and 35 per cent of the "Hungaro-Frisian" ones. Further, the data make it clear that it is first of all in the poor and medium intensive symptoms of oestrus that the genotypes show significant differences. Also, it follows from the observations that the oestrus is most difficult to detect in the Hungarian spotted breed; as, under bound keeping conditions, 34 per cent of these cows display very poor or poor symptoms of oestrus (Fig. 2).

In Fig. 2 the time spent by then Holstein-Frisian cows in lying and activity (moving, jumping), respectively, before and during the oestrus is seen as percentages of 24 hours. The time of lying grew from 15 to 48 per cent.

According to the observations at the time of feeding, cows in heat went also to the trough and jumping hardly occurred. So then, in a bound keeping system animals in heat are very difficult to detect. Only 5.34 per cent of the whole time of jumping fell to the daily two periods of feeding.

The daily distribution of jumping about of cows in heat is shown in Fig. 3. The larger part (some 65 per cent) of the jumping activity fell between 9 p.m. and 4 a.m. Thus, observations of cows in heat under bound keeping conditions give the best results when made in this period.

Similar conclusions were drawn by SAMBRAUS (1978) who found that the milking breeds were no more active during the oestrus than the dual-purpose ones.

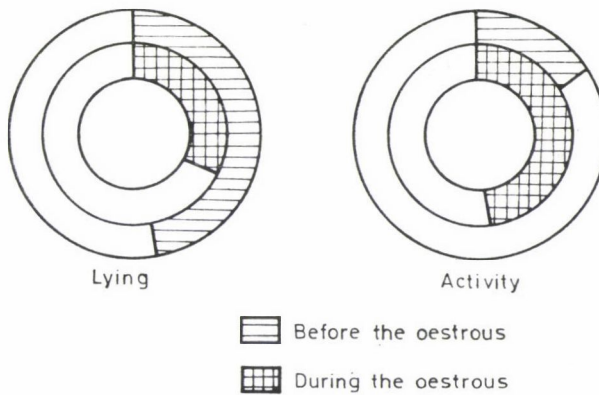


Fig. 2. Behaviour of cows in heat before and during the oestrus under free-pen conditions

Another observation we made was that the frequency of jumping depended upon the behaviour of both cows in heat and those being jumped upon. One of the cows in heat jumped on 84 occasions upon 19 cows. Another cow jumped on 42 occasions upon 11 cows, while a third one did so on 5 occasions upon 4 cows. Considering that we could not watch all cows during the oestrus we have no data as to whether or not the frequency of jumping about was in relation with the place occupied by the cow in the hierarchy of the group. Namely, the authors do not agree on this point. According to KILGOUR (1979) cows superior in rank jump more frequently, while HURNIK and KING (1979) found that though the frequency of jumping did show correlation with the hierarchy, this correlation was hardly significant. So the question requires further investigations, all the more so because the data known from the literature refer to keeping conditions different from those in our study.

Correlations between the length of the period of oestrus and the standards of different qualities are summarized in Table 2. According to the data of the table, the length of the period of oestrus was found to be closest correlated with the body weight. The lower

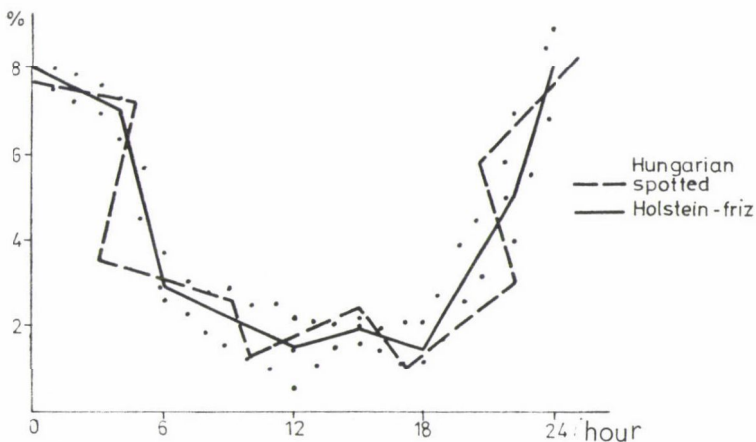


Fig. 3. Daily distribution of the jumping activity of cows in heat



Table 2

*Correlations between the length of the period of oestrus and the individual standard qualities*

Designation	Breed	<i>r</i>	P%
Oestrus period — body weight	Hungarian red spotted	—0.46	1.0
	Holstein-Frisian	—0.29	1.0
	"Hungaro-Frisian"	—0.84	0.1
Oestrus period — 100-day milk production	Holstein-Frisian	—0.16	10.0
	"Hungaro-Frisian"	—0.28	5.0
Oestrus period — calving interval	Hungarian red spotted	—0.23	5.0
	Holstein-Frisian	—0.36	5.0
	"Hungaro-Frisian"	—0.52	0.1
Number of jumpings per oestrous cycle — pregnancy index	Hungarian red spotted	+0.75	1.0

the body weight, the longer the period of oestrus, which suggests that any increase in the body weight reduces the time of oestrus. The correlation was the most expressed in the "Hungaro-Frisian" stock.

Between the length of the oestrous period and the 100-day milk production, a poor negative correlation was obtained. The correlation of the oestrus period with the time interval between calvings was weak and medium negative, but significant. These values indicate that cows in heat for a longer time are easier to notice and consequently earlier inseminated. Furthermore, these correlations point to the fact that the qualities greatly depend on environmental factors.

The significant correlation between the frequency of jumping per oestrous cycle and the index of pregnancy suggests that cows exhibiting intensive activity during the oestrus are more reliably inseminated. Since activity during the oestrus is nearly the same in milking and dual-purpose breeds, according to the literature and our own experiences alike, the correlation between oestrous activity and conception can be regarded as a fact.

The conclusion drawn from the investigations completed is that, while the length of the period of oestrus is not influenced by the keeping system (bound or free-pen), there is a considerable difference between milking- and dual-purpose types. Thus, the detection of cows in heat is more difficult in the Hungarian red spotted breed than in the Holstein-Frisian or "Hungaro-Frisian" populations which show the symptoms of oestrus for a longer time. In addition, under bound keeping conditions more than one-third of the Hungarian red spotted cows exhibit poor or very poor symptoms of oestrus.

At the same time, in the free-pen system the activity of Hungarian spotted cows in heat corresponds to that of the Holstein-Frisian breed. Since the data obtained in the experiment can be generalized, the idea arises that under industry-like free-pen conditions the Hungarian red spotted breed will also show more favourable reproduction indexes than in the present system of bound keeping.

A new result of our observations is that, in the free-pen system of keeping, animals in heat cannot be detected at the time of feeding; namely, the earlier views that the feeding time of cows in heat is considerably reduced, and that they jump on each other, need revision.

The correlations between the length of the period of oestrus and the standard qualities suggest that while a selection for populations showing symptoms of oestrus for a longer time

is favourable in a keeping system of industrial character, the detection of these symptoms depends primarily on the environmental conditions.

On the basis of our experiments and investigations our proposals for the practice are:

a) Since in 70–80 per cent of the cows the first symptoms of oestrus appear in the small hours, animals showing such symptoms in the morning or before noon have to be regarded as if the oestrus had begun at dawn.

b) Under free-pen conditions, cows not lying but standing apart at night and dawn must also be examined besides those jumping about. In this way differences between the free-pen and the bound system of keeping in the early detection of the oestrous symptoms might be considerably reduced.

c) A considerable proportion of the activities of jumping about and standing apart, related with the oestrus, occurs at dawn and in the early morning hours. Therefore, if not enough time is available for the selection of cows in heat, this work had best be done in those hours.

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### References

- ANDREAE, U. and PFLEIDERER, U. E. (1977): Über die Aussagefähigkeit von Brunstbeobachtungen am Rind. *Der Tierzüchter*, **29**, 196–199.
- CZAKÓ, J. (1978): *Gazdasági állatok viselkedése* (Behaviour of farm animals). Mezőgazdasági Kiadó, Budapest.
- HURNIK, J. F. and KING, G. J. (1979): A holstein-friz tehének ivarzási viselkedése ellés után kötetlen istállóban (Manifestation of oestrus in Holstein-Frisian cows after calving in free-pen system of keeping). *ÁKI Kiadvány, Herceghalom*, 27–34.
- KILGOUR, R. (1979): Einige für Neuseeland wichtige Aspekte der angewandten Verhaltensforschung. *Der Tierzüchter*, Hildesheim, **31**, 99–101.
- KING, G. J. and HURNIK, J. F. (1979): A reprodukció és az ivarzási tünetek megjelenése az ellés után (Appearance of reproductive function and oestrous symptoms after calving). *ÁKI Kiadvány, Herceghalom*, 37–44.
- MYLREA, P. J. and BEILHARZ, R. G. (1964): The manifestation and detection of oestrus in heifers. *Animal Behaviour*, **12**/1, 25–30.
- SAMBRAUS, H. H. (1973): *Das Sexualverhalten der Wiederkäuer*. Forscher, Verhaltensforsch. H. 12. Paul Parey, Berlin—Hamburg.
- SAMBRAUS, H. H. (1978): *Nutztier Ethologie*. Paul Parey, Berlin—Hamburg.

### EFFECT OF UREA ON PROTEIN METABOLISM OF ANGORA RABBITS

The Rabbit's caecum is analogous with the forestomachs of ruminants in some of its functions, including decomposition of cellulose, transformation of protein, synthesis of vitamin B, etc. Endogenous urea is also utilized for bacterial protein synthesis in the caecum (HÖRNICKE 1972). Bacterial proteins are rich in essential amino acids. If free amino acids are liberated, they are absorbed from the caecum, but most of the protein can only be utilized after consumption and digestion of the soft (caecal) excrement.

However, in respect of the importance of caecal function there is disagreement among the various authors. ALEKSIEV (1963) obtained negative nitrogen balance when he fed urea to rabbits on 0.6–2.0% level. According to KING (1971) NPN utilization is negligible if the protein content of the ration is higher than 10%, i.e., if it exceeds the requirement for maintenance. NIEDZWIADK and KAWINSKA (1975) showed that weight gain of the rabbits was not affected by urea, if it substituted for half of the 9% fish meal in the ration. LEBAS and COLIN (1973) could not demonstrate a significant effect of urea by supplementing with it a diet of 12.5% protein content. HOUP (1963) found that caecal microorganisms synthesize a considerable amount of bacterial protein from urea in the rabbit. Urea is excreted across the intestinal mucosa into the lumen of the digestive tract where it is hydrolysed becomes ammonia and CO<sub>2</sub>. The ammonia produced is partly absorbed. In protein deficient rabbits, the proportion of endogenous urea utilized for protein synthesis may reach 40%.

CHEEKE (1972) and CHEEKE and AMBERG (1972) suggested that in the caecum, bacterial-protein synthesis is not independent from the dietary supply of essential amino acids. According to SCHLOLAUT (sine anno) the conversion of NPN is improved when the feed is supplemented with essential amino acids.

In earlier experiments (TELEKI *et al.* 1981) we have shown that by the end of the production period (before shearing) serum urea and ammonia levels rose to a considerable extent in Angora rabbits, suggesting significant protein catabolism. On the other hand, the level of some amino acids, especially that of methionine, decreased.

In the present work we investigated whether Angora rabbits are able to utilize urea and if they can whether this is affected by the supplementation of essential amino acids.

Ten fully developed, approximately two-year old male Angora rabbits were divided in two groups and placed in individual metabolic cages. The mean initial live weights were  $3.18 \pm 0.30$  and  $3.22 \pm 0.26$  kg in the control and experimental group, respectively.

The daily ration of each group was 120 g granulated feed mixture of 5 mm diameter given in the morning. The feed of the control group contained 16.82% crude protein, that of the experimental group 16.53%. Most of the crude protein was true protein, 14.02% of the dry matter. In the experimental group 15.8% of crude protein was substituted by urea-N. The amount of other nutrients in the ration was nearly the same in the two groups (Table 1).

Table 1

*Nutrient composition of feeds*

Groups	Dry matter	Crude ash	Crude protein	Protein	Crude fat	Crude fibre	N-free extract
Control	91.0	5.64	16.82	—	3.11	10.26	55.17
Urea feed	91.0	4.55	16.53	14.02	1.85	10.67	57.40

Faeces and urine was collected daily. The samples were not pooled. Feed residues also were collected daily and weighed.

The experiment consisted of a seven-day adaptation period and a five-day experimental period. On the last day, blood samples were taken from the ear vein for chemical analyses.

Serum protein was determined with the biuret reaction, ammonia by the method of KELLER *et al.* (1967), urea after FAWCET and SCOTT (1960), amino-N with beta-naphthochinon-4-sulfonic acid and haemoglobin with the DRABKIN procedure as modified by ZILJSTRA and



KAMPEN (1960). Blood sugar was determined with the orthotoluidine technique, lactic acid by the method of VELŐSY (1979), cholesterol according to WATSON (1960).  $\text{Ca}^{2+}$  was measured by flame photometry, and inorganic P with the molybdenum blue reaction.

*Nitrogen balance.* The experimental rabbits consumed less feed and therefore less nitrogen, than did the controls, probably due to the presence of urea (Tables 2 and 3).

Table 2

*N-balance data of experimental and control animals  
(N g/animal/day)*

Groups	Control	Urea feed
Total N consumed	$3.177 \pm 0.05$	$2.962 \pm 0.06$
Urea N	—	$0.475 \pm 0.01$
Protein N	3.177	2.487
Faecal N	$0.518 \pm 0.14$	$0.501 \pm 0.08$
Urinary N	$1.459 \pm 0.50$	$1.775 \pm 0.32$
Retained N	$1.199 \pm 0.45$	$0.687 \pm 0.31$

Table 3

*N-balance data as percentages to total N consumed*

Groups	Control	Urea feed
Faecal N	16.32	16.92
Urinary N	45.91	59.91
Nitrogen retention	37.76	23.28
Retention of protein N	—	27.58

Urinary N excretion was significantly higher in the urea-group ( $P < 0.05$ ). As a consequence, the N retention of the control group surpassed that of the experimental rabbits (Tables 2–4). Rabbits of both groups gained about 30–40 g in the experimental period.

*Chemical analyses.* Total serum protein ( $P < 0.1$ ), haemoglobin ( $P < 0.05$ ) and cholesterol ( $P < 0.05$ ) levels were significantly lower in the urea group than in the experimental rabbits, while the opposite was true for ammonia ( $P < 0.05$ ), urea ( $P < 0.05$ ) and  $\text{Ca}^{2+}$  ( $P < 0.05$ ), as is shown in Table 5. No significant differences were found in blood sugar lactic acid and inorganic P (Table 5).

Animals given urea excreted significantly more nitrogen with the urine than the control rabbits; in accordance, the utilization of nitrogen was less in the group consuming urea. It is known that if less protein is fed, its utilization will not be impaired, and in most

**Table 4**

*N-balance data per kg body weight*  
(N g/day/kg body weight)

Groups	Control	Urea feed
N consumed	0.999	0.919
Faecal N	0.163	0.156
Urinary N	0.459	0.551
N retention	0.377	0.213
Urea N consumed	—	0.147

**Table 5**

*Chemical analyses*

Groups	Control	Urea feed
Serum protein, g/l	70.30 $\pm$ 3.6	65.80 $\pm$ 2.7
Ammonia, mmol/l	119.50 $\pm$ 10.1	176.00 $\pm$ 10.0
Urea, mmol/l	6.73 $\pm$ 0.9	9.75 $\pm$ 1.6
Amino acid N, mmol/l	7.98 $\pm$ 0.7	8.83 $\pm$ 1.2
Haemoglobin (Fe), mmol/l	5.80 $\pm$ 0.7	4.80 $\pm$ 0.4
Blood sugar, mmol/l	4.30 $\pm$ 0.3	4.20 $\pm$ 0.6
Total lactic acid, mmol/l	8.97 $\pm$ 2.2	9.30 $\pm$ 1.3
Total cholesterine, mmol/l	1.90 $\pm$ 0.44	1.25 $\pm$ 0.17
Inorganic P, mmol/l	1.12 $\pm$ 0.06	1.24 $\pm$ 0.1
Ca <sup>++</sup> , mmol/l	3.03 $\pm$ 0.31	3.14 $\pm$ 0.09

cases it is even improved. Therefore the lower utilization of protein-N in the urea group may not be due to the somewhat reduced protein content of the feed. Thus, the conclusion can be drawn that dietary urea was not utilized by the rabbits. Moreover, urea even decreased the utilization of protein by some 10%. Results of chemical determinations support the conclusions drawn from the N-balance experiments. The low serum protein and haemoglobin levels and high ammonia and urea levels in the blood of experimental rabbits indicate the possibility of a latent ammonia intoxication.

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## References

- ALEKSIEV, A. D. (1963): Ispolzuvane na karbamid v hraneneto na zajcite Naucs. Tr. Szofia, **13**, 279.
- CHEEKE, P. R. and AMBERG, J. W. (1972): Protein nutrition of the rabbit. *Nutr. Rpts.*, **5**, 259.
- CHEEKE, P. R. (1972): Nutrient requirements of the rabbit. *Feedstuffs*, **44**, 28.
- FAWCETT, J. K. and SCOTT, J. E. (1960): A rapid and precise method for the determination of urea. *J. Clin. Path.*, **13**, 156.
- HOUP, T. R. (1963): Urea utilization by rabbits fed a low-protein ration. *Am. J. Physiol.*, **205/6**, 1144.
- HÖRNICKE, H. (1972): Neuere Befunde über Blinddarm-funktionen und Coecotrophie bei Kaninchen. *Z. Tierphysiol. Tierernährung u. Futtermittelkunde*, **30**, 121.
- KELLER, H., MÜLLER-BEISENHIRTZ, W. and NEUMANN, E. (1976): Eine Methode zur Ammoniakbestimmung in Kapillarblut. *Klin. Wschr.*, **45**, 314.
- KING, J. O. L. (1971): Urea as a protein supplement of growing rabbits. *British Veterinary Journal*, **127**, 523.
- LEBAS, F. and COLIN, M. (1973): Effect de l'addition de l'urée à un régime pauvre en protéines chez le lapin en croissance. *Ann. Zootechn.*, **22**, 111.
- NIEDZWIADK, S. and KAWINSKA, J. (1976): Untersuchungen über den Harnstoffeinsatz in der Kaninchen-fütterung. *L. Cong. Int. Cunic. Dijon*.
- SCHLOLAUT, W.: Die Ernährung des Kaninchens. Hoffmann La-Roche A.G. 1983. Grenzach-Wyhlen.
- SZILÁGYI, L. (1971): Módszergyűjtemény orv. laboratóriumok számára (Collection of methods for medical laboratories). Magyar Optikai Művek, Budapest, 1982.
- TELEKI, M., SZEGEDI, B. and JÉCSAI, J. (1981): Különböző összetételű takarmánykeverékek hatása a termelő angoranyulak gyapjúhozamára, fehérje-, aminosav és ásványanyag-forgalmára. (Effect of various compositions of feed mixture on the wool yield and on the protein, amino acid and mineral metabolism of productive Angora rabbits). *Állattenyésztés, Takarmányozás* (in press). ÁTK. Közleményei, Herceghalom.
- VELŐSY, GY. (1979): Tejsav- (laktát) meghatározás fotometriás mikromódszerrel. [Determination of lactic acid (lactate) by photometric micromethod]. *Kísérletes Orvostudomány*, **31**, 662.
- WATTSON, D. (1960): A simple method for the determination of serum cholesterol. *Clin. Chem. Acta*, **5**, 637.
- ZILJSTRA, W. G. and KAMPEN, E. J. (1960): Standardization of hemoglobinometry. I. The extinction coefficient of hemoglobincyanide. *Clin. Chem. Acta*, **5**, 719.

## STUDIES ON THE QUALITY OF RABBIT MEAT II—PHYSICAL AND HISTOLOGICAL CHARACTERISTICS

The meat quality of large animals and different kinds of poultry was studied extensively (HOWARD and LAWRIE 1956, HAMM 1960, CARPENTER 1962, BLUMER 1963, BRAY 1964, GROU and HAMM 1964 and KAUFFMAN *et al.* 1969 on bovine and porcine meat and DODGE and STADELMAN 1960, KLOSE *et al.* 1960, SPENCER *et al.* 1961 and others on different kinds of poultry meat). However, the quality of rabbit meat had not yet received as much attention. The object of the present work is to study the effect of age, sex and kind of muscles on the quality of rabbit meat as measured by physical and histological characteristics.

This research was carried out at Animal and Poultry Production Department, Faculty of Agriculture, Assiut University. Details about the number, age and management of experimental animals, as well as processing and sampling, were as described by EL-GAMMAL *et al.* (1980). The expressible fluid was determined by applying the method of BAKER *et al.* (1972).

Samples of muscles from the hind limb (*Rectus femoris m.*), fore limb (*Triceps brachii m.*) and loin (*Psoas major m.*) were directly fixed in 110 per cent formalin solution. After fixation the samples were dehydrated, cleared and embedded in paraffin wax. Paraffin sections (6 microns thickness) were taken, stained by eosin-haematoxyline and microscopically examined to determine the diameter and number of fibers per square centimeter of muscle bundle. Then,



both fiber and connective tissue areas were calculated and expressed as percentages of total muscular tissues.

Statistical analysis was done applying the methods of SNEDECOR and COCHRAN (1967).

In the present study the quality of rabbit meat was determined physically by measuring expressible fluid, which reflects the water-holding capacity of meat (the higher the expressible fluid, the lower the water holding capacity) and histologically by measuring fiber diameter as well as counting their number in 1 cm<sup>2</sup> of muscle tissues.

### *I. Physical characteristics*

#### *Expressible fluid:*

Water in fresh meat represents about 75%. Thus, it influences both the qualities of juiciness and tenderness. HAMM (1960) defined water-holding capacity as the ability of meat to retain its own or added water during the application of any force such as pressing, heating, chilling or grinding. Water liberated by such methods is termed as free or loose water, and water that remains is called bound water. Since loose water, usually obtained by pressure, contains some soluble constituents, it may be called expressible fluid.

The expressible fluid of rabbit meat decreases with advancing age till maturity. At one month of age, expressible fluid is 170–269 mg per gram of meat and decreases to 46–62 mg at 9 months of age. This means that water-holding capacity increases with advancing age. SCHON and SCHEPER (1960) found significantly less expressible juice in veal than in beef. The decrease in expressible fluid with age may relate to the decrease in total moisture content, especially extracellular water. EL-GAMMAL *et al.* (1980) found that the total moisture content of rabbit meat decreased with advancing age. However, EL-GAMMAL (1977) in his work on the water-holding capacity of chicken meat, indicated that the total moisture content of meat is not the only factor determining the expressible fluid but that other factors may be involved.

Analysis of variance (Table 2) shows that the effect of age on the expressible fluid of rabbit meat has a highly significant linear trend, but that of the quadratic is insignificant.

In regard to gender, Table 1 indicates that, in most cases female meat has higher values of expressible fluids than that of males, especially in the loin and fore limb parts. This may be due to differences in chemical composition (EL-GAMMAL 1977). Other factors may be involved, such as differences in pH (HAMM 1960); in mineral content of muscles (ASSAF and BRATZLER 1966) or in the kind of ion charges on protein surface (HAMM 1955).

**Table 1**  
*Expressible fluid\* of rabbit meat as affected by age and sex*

Age (months)	Fore limb		Loin		Hind limb	
	male	female	male	female	male	female
1	170	227	109	197	269	260
2	153	210	163	174	182	141
3	126	148	119	101	137	134
4	123	80	76	117	118	128
5	87	87	76	97	84	74
6	73	94	71	75	78	70
7	65	68	68	73	55	69
8	58	66	68	69	54	48
9	48	58	62	60	49	46

\* Expressed as milligrams per one gram of meat.

It is quite obvious from the analysis of variance (Table 2) that the differences due to gender are insignificant.

The expressible fluid differed according to the kind of muscles. During the first three months, the hind limb muscles had more expressible fluid than those of the other cut-up

Table 2

*Analysis of variance of expressable fluid of rabbit as influenced by age, sex and cut-up parts of the carcass*

Source of variation	d.f.	S.S.	M.S.	F
Age (A)	8	2677.41	334.67	7.57**
Linear	1	2375.90	2375.90	53.74**
Quadratic	1	26.43	26.43	0.59
Cubic	1	9.70	9.70	0.21
Deviation	5	265.38	53.07	1.20
Part (P)	2	29.42	14.71	0.33
Sex (S)	1	19.59	19.59	0.44
A × P	16	280.66	17.54	0.39
A × D	8	47.98	5.99	0.13
P × S	2	39.51	19.75	0.44
Error	70	3094.55	44.20	

\* Significant at P<sub>5</sub>% level.

\*\* Significant at P<sub>1</sub>% level.

parts. However, it is difficult to detect such differences at the later ages. In general, the analysis of variance showed that the differences in the expressible fluid of different cut-up parts are insignificant (Table 2).

## II. Histological characteristics

### 1. Number of muscle fibers per one square centimeter

As shown in Table 3, the number of muscle fibers in an area unit decreases as the rabbit ages. This decrease ranges from about 20 to 45% according to type of muscles and sex. The decrease in fiber number by age is associated with the increase in fiber diameter (see Table 5). However, the area resulting from the increase in fiber diameter by age surpasses the area resulting from the decrease in fiber number by age KAWEKA (1963) and ABERLE (1978) in their work on chicken meat, found that the number of muscle fibers in an area unit decreased and their diameter increased as the birds grew older.

Analysis of variance (Table 4) shows that the effect of age on the number of muscle fibers has a highly significant linear trend, while those of quadratic or cubic sources of variation are insignificant.

As for gender, it appears that female muscles have higher number of muscle fibers per area unit than those of males.

The comparison of muscle fibers in different muscles shows that the loin muscles have the highest number of muscle fibers, followed by those of the hind limb and, finally, those of the fore limb.

Table 3

*Number of muscle fibers in one square centimeter as affected by age, gender and cut-up parts of rabbit carcasses*

Age (months)	Fore limb		Loin		Hind limb	
	male	female	male	female	male	female
at birth	34 640	41 102	39 307	56 512	38 474	46 230
1	33 153	40 794	37 230	54 179	38 025	43 473
2	30 730	38 217	36 243	52 499	35 836	42 384
3	27 503	31 153	35 422	41 666	33 333	42 128
4	25 128	30 191	34 589	41 025	31 794	41 358
5	25 179	30 871	34 063	41 128	29 461	36 204
6	24 499	30 794	32 064	33 820	28 909	35 717
7	22 128	30 294	30 140	33 461	26 769	33 256
8	20 769	29 435	29 858	33 384	25 422	31 063
9	20 717	21 794	29 179	33 371	22 191	23 409

Analysis of variance (Table 4) indicates highly significant differences in the number of muscle fibers of different cut-up parts. However, the differences due to gender are insignificant.

Table 4

*Analysis of variance of the number of muscle fibers in an area unit as influenced by age, gender and cut-up parts of carcass*

Source of variation	d.f.	S.S.	M.S.	F
Age	9	3 663 983 500	407 109 277	10.11**
Linear	1	3 597 586 515	3 597 586 515	89.38**
Quadratic	1	8 949 343	8 949 343	0.22
Cubic	1	3 775 978	3 775 978	0.09
Deviation	6	53 671 664	8 945 277	0.22
Part	2	1 537 465 600	768 732 800	19.09**
Sex	1	75 706 500	75 706 500	1.88
Error	107	4 306 536 300	40 248 002	

\* Significant at P<sub>5%</sub> level.

\*\* Significant at P<sub>1%</sub> level.

## 2. Diameter of muscle fibers

The diameter of muscle fibers of different cut-up parts of both males and females are presented in Table 5.

The diameter of muscle fiber increased with the animal's growth to the end of the experimental period. Irrespective of sex and kind of cut-up parts, the fiber diameter ranged from about 20–28 microns at one month of age and reached about 46–52 microns at nine months. In other words, the fiber diameter increased about 90–130% over eight months.

However, it is difficult to detect sex difference in fiber diameter to four months of age; but after this age, the female muscle fibers are of slightly greater diameter than male muscle fibers.



**Table 5**  
*Fiber diameter (Microne) as affected by age, gender  
 and kind of muscle*

Age (months)	Fore limb		Loin		Hind limb	
	male	female	male	female	male	female
1	27.5	28.6	22.5	22.5	20.4	20.9
2	28.6	29.2	29.2	29.7	29.2	28.6
3	29.2	29.3	33.0	34.1	29.7	28.9
4	39.6	39.6	40.2	40.1	33.5	33.0
5	40.2	41.1	40.5	40.9	39.0	39.6
6	41.8	42.8	41.5	43.7	42.1	43.1
7	43.7	44.2	43.5	45.1	44.0	45.0
8	44.5	44.9	44.0	46.7	45.6	46.2
9	50.1	51.7	51.1	51.7	46.7	46.9

Table 5 shows that at one month of age the diameter of fore limb muscle fibers is greater than that of other muscle fibers, and those of the hind limb have the least fiber diameter. After one month of age until full maturity, the loin muscle fibers are of the greatest diameter. The fore limb muscle fibers are of greater diameter than those of hind limb muscles until four months of age, after which the latter becomes greater.

**Table 6**  
*Analysis of variance of fiber diameter as affected  
 by age, gender and kind of muscle*

Source of variation	d.f.	S.S.	M.S.	F
Age	8	3511.5	438.9	71.7**
Muscle	2	35.9	179	2.9
Sex	1	1.9	1.9	0.32
Error	42	257.2	6.2	

Statistical analysis (Table 6) shows that differences due to age are highly significant while those due to type of muscle or gender are insignificant.

### 3. Fiber and connective tissue areas

The calculated values of connective and fiber areas of different muscles are shown in Table 7.

Irrespective of gender and type of muscle, fiber area percentage increases by age from 12.4–26.2% at one month to reach 37.9–70.1% at nine months. Looking again at Tables 3 and 5, it can be seen that the number of fibers decreases while fiber diameter increases with age. This means that the fiber diameter is more important than fiber number in determining fiber area percentage. On the other hand, the connective tissue area percentage decreases with age. KHALIFA (1980), working on duck muscles, came to the same conclusion. WILSON *et al.* (1954) reported that older animals did not contain greater quantities of con-

nective tissues than younger animals. Although the connective tissue percentage decreased by age, it became tougher (CARMICHAEL and LOWRIE 1967) and this may be the reason for decrease of tenderness with age. In other words, the use of connective tissues percentage for evaluating tenderness of meat at different ages will be contradictory. However, it will be of importance when used to compare tenderness between different muscles of the same animal, or different animals of the same breed and age.

As shown from Table 7 the female muscles have comparatively greater content of fiber tissue than do those of the male. This means that the male muscles have a greater content of connective tissue than do female muscles.

**Table 7**

*Fiber and connective tissue areas (%) in Bouscat rabbit muscles as affected by age and gender*

Age (months)	Fore limb				Loin				Hind limb			
	male		female		male		female		male		female	
	A	B	A	B	A	B	A	B	A	B	A	B
1	19.9	80.1	26.2	73.3	14.8	85.2	21.5	78.5	12.4	87.6	14.8	85.1
2	19.7	80.3	25.6	74.4	24.3	75.7	36.3	63.7	23.9	76.1	27.2	72.8
3	18.4	81.6	20.8	79.2	30.3	69.7	38.6	62.0	29.7	70.3	27.6	72.4
4	30.9	69.1	37.1	62.9	43.8	56.2	51.7	48.3	28.0	72.0	35.4	64.6
5	31.9	68.1	40.9	58.1	43.8	56.2	54.0	46.0	35.2	64.8	44.6	55.4
6	33.6	66.4	44.3	55.7	43.4	56.6	56.7	43.3	40.2	59.8	52.1	47.9
7	33.1	66.9	46.5	53.5	44.8	55.2	53.4	46.6	40.7	59.3	52.8	47.1
8	32.2	67.8	46.5	53.5	45.4	54.6	57.2	42.8	41.5	58.5	52.1	47.9
9	40.8	59.2	45.6	54.3	59.8	40.2	70.1	29.9	37.9	62.1	40.4	59.6

A = Fiber tissue area (%).

B = Connective tissue area (%).

The loin muscles have a lesser content of connective tissue than do other muscles. However, it is difficult to detect such difference between fore and hind limb muscles because in some cases the former had more connective tissue content than did the latter, while the opposite was true in the other cases.

KHALIFA (1980), in his work on duck meat, concluded that protein content of muscles increases as the fiber area increases while fat content decreases as the connective tissue area decreases.

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### References

- ABERLE, E. D., ADDIS, P. B. and SHOFFNER, R. N. (1978): Fiber types in skeletal muscles of broiler and layer type chickens. *Poultry Sci.*, **58**, 1210.  
 ASSAF, S. A. and BRATZLER, L. J. (1966): Inorganic elements in beef muscle and their relative degree of binding in aqueous beef muscle extracts. *J. of Agric. and food chemistry*, **14**, 487.

- BAKER, R. C., DARFLER, J. M. and ORTLIEB, W. (1972): The effect of rate of cooking on the quality of the Barbecued chicken. *Poultry Sci.*, **51**, 5: 1656.
- BLUMER, T. N. (1963): Relationship of marbling to the palatability of beef. *J. Animal Sci.*, **22**, 771.
- BRAY, R. W. (1964): A special study of the beef grade standards Proc. 17th. Reciprocal Meat conf. 1964.
- BRISKY, E. J. and LISTER, D. (1968): Influence of stress on Pork muscle quality. Future for Pork conference (Iowa state Univ., Ames).
- CARMICHAEL, D. J. and LOWRIE, R. A. (1967): Bovine Collagen: 1 — Changes in collagen solubility with animal age. *J. Food Tech.*, **2**, 299.
- CARPENTER, Z. L. (1962): The histological and physical characteristics of pork muscle and their relationship to quality. Ph.D. thesis, Univ. of Wisconsin. Cited by Price and Schweighert, 1970.
- DODGE, J. W. and STADELMAN, W. J. (1960): Studies on tenderness evaluation. *Poultry Sci.*, **39**, 134.
- EL-GAMMAL, A. M. (1977): Water binding capacity of chicken meat and its relation to chemical composition and storage period. Accepted for publication in *Assiut J. of Agric. Sci.* (in press).
- EL-GAMMAL, A. M., MAKLED, M. N. and ABDEL-NABY, M. A. (1980): Studies on the quality of rabbit meat. 1 — Chemical characteristics, (under publication).
- GRAU, R. and HAMM, R. (1953): A simple method for determination of water-holding capacity in muscles. *Naturwissenschaften*, **40**, 29.
- HAMM, R. (1955): Die Ursache der Wirkung von Bratzusatzmitteln und Kochsalz auf Fleisch. *Die Fleischwirtschaft*, April, 196–205.
- HAMM, R. B. (1960): Biochemistry and meat hydration. *Advance Food Res.*, **10**, 355.
- HOWARD, A. and LOWERIE, R. A. (1956): Studies on beef quality. II. Physiological and biological effects of various pre slaughter treatment. *Food, Preserv. Transp. Tech. Paper*, **2**, 18.
- KAUFFMAN, R. C., KALB, Q. E., BREIDEN STEIN, B. C. and CARRIGAN, D. S. (1969): Meat quality. Univ. II coop ext. serv. Circ. 1007. Cited by Price and Schweighert, 1970.
- KAWEKA, B. M. (1963): Comparison of the growth, histological structure and chemical composition of the meat of crossbred pullets and pullets of the parental breeds. *A.B.A.* Vol. **33**, 2: 302.
- KHALIFA, R. M. (1980): Studies on some reproductive traits in Peking ducks. M.Sc. thesis, Faculty of Agriculture, Minia University.
- KLOSE, A. A., CAMPBELL, A. A. and HAUSAN, H. L. (1960): Stability of frozen ready-to-cook chicks. *Poultry Sci.*, **39**, 1136.
- PRICE, J. F. and SCHWEIGHERT, B. S. (1970): The science of meat and meat products. Second edition. W. H. Freeman and company. San Francisco.
- SCHON, L. and SCHEPER, J. (1960): Merkmale der Beschaffenheit von Schweine-, Kalb- und Rindfleisch und ihre Beziehungen, zueinander unter dem Gesichtspunkt verschiedener Verwahrungsformen. *Züchtungskunde*, **32**, 788.
- SNEDECOR, G. W. and COCHRAN, W. C. (1967): Statistical methods. 6th edition. Calcutta, Bombay, New Delhi, Oxford and BH Publishing companies.
- SPENCER, J. V., SAUTER, E. A. and STADELMAN, W. J. (1961): Effect of freezing, thawing and storage on spoilage flavour and bone darkening. *Poultry Sci.*, **40**, 918.
- WILSON, G. D., BRAY, R. W. and PHILLIPS, PH. H. (1954): The effect of age and grade on the collagen and elastin content of beef and veal. *J. Animal Sci.*, **13**, 826.





## LECTIONES

### CHARACTERIZATION AND ASSESSMENT OF SOIL SALINITY AND ALKALINITY

When speaking of the distribution of saline and alkali soils in different areas, we have to consider those geochemical features of the landscape which influence the development of salt-affected soils.

Roughly one tenth of all dry land is covered by saline and alkali soils. However their distribution is uneven — in some regions they occur either seldom or not at all, whereas in other regions they are very common and sometimes dominant.

#### *Salt accumulation in different landscapes*

It is known, that arid and semi-arid conditions contribute to salt accumulation in soils and waters and, as a result, the formation of salt-affected soils in desert and semi-desert regions is quite a common phenomenon. The accumulation of electrolytes in soils is, however, a complex process which cannot be interpreted as a simple consequence of climatic effects. The hydrology as well as hydrogeology and geomorphology of a given area have substantial influence on salt migration in waters and grounds. The integrated effects of environmental factors determine the trend of salt migration and the balance of soluble products accumulating in soil layers.

Based on the landscape geochemistry studies of KOVDA (1946-47), POLINOV (1956) and SZABOLCS (1981), the following landscape types can be distinguished as geochemical regions of salt accumulation in soils.

**Table 1**  
*Scheme of landscape geochemistry of salt-affected soils*

Landscape	Main chemical type of salt accumulation	Dominant type of salt-affected soils
Salt dome	NaCl	solonchak
Maritime	NaCl	solonchak
Deluvial	Na <sub>2</sub> SO <sub>4</sub> , NaHCO <sub>3</sub>	solonchak, solonetz
River deltas	NaCl, Na <sub>2</sub> SO <sub>4</sub> , NaHCO <sub>3</sub> , Na <sub>2</sub> CO <sub>3</sub>	solonchak
Dry deltas	NaCl, Na <sub>2</sub> SO <sub>4</sub>	solonchak, solonetz
Alluvial plains	Na <sub>2</sub> SO <sub>4</sub> , NaCl, NaHCO <sub>3</sub> , Na <sub>2</sub> CO <sub>3</sub>	solonchak, solonetz
River terraces	NaHCO <sub>3</sub> , Na <sub>2</sub> CO <sub>3</sub> , Na <sub>2</sub> SO <sub>4</sub> , NaCl	solonetz
Salt-lakes and marches or swamps	NaCl, Na <sub>2</sub> SO <sub>4</sub> , Na <sub>2</sub> CO <sub>3</sub>	solonchak

\* Lecture held at the Fifth International Soil Classification Workshop, Sudan, 2-11. Nov. 1982.

In Table 1 the landscape types are listed, in which salt accumulation occurs and salt-affected soils may develop. As can be seen in the Table, different landscape types are associated with different chemical and pedological types of salt accumulation as well as with different soil types. In several landscape types, the climatic conditions have substantial influence on the chemistry and magnitude of salinity and alkalinity. On alluvial plains, for instance, in arid regions  $\text{Na}_2\text{SO}_4$  and  $\text{NaCl}$  accumulate, resulting in solonchak soils; while in more humid areas  $\text{Na}_2\text{CO}_3$ — $\text{NaHCO}_3$  accumulation prevails, associated with solonetz soils.

We must agree that saline and alkali soils develop in different climatical as well as geochemical regions influencing the genetics and properties of many soil types. In the legend of the Soil Map of the World and in Soil Taxonomy the term "Saline phase" or "Sodic phase" indicates the occurrence of salt accumulation in different soils. If, due to natural or antropogen factors in soil formation the accumulation of salts is progressive, from the given soil in saline phase — or alkali phase, solonchak or solonetz may develop.

#### *Influence of salinity and alkalinity on different soils*

As it is clear from the foregoing, salt accumulation in soils is always associated with certain environmental conditions. Given the climatical, hydrological and geomorphological properties of the landscape, the soil material, whose composition and pattern we express as type, or in other taxonomical terms in our classification systems, is primarily exposed to the influence of increasing or decreasing salt concentration in the soil solution. As a result of interaction of salts with soil material, different kinds of salt-affected soils develop. For this reason, some salt-affected soils dominate in a variety of soil regions. For example, it is well known that, in the environment of arid soils, solonchaks are frequent and no solonetz can be found in deserts and semi-deserts. On the other hand, in alluvial plains and river terraces of moderate climatical areas, solonetz is the prevalent type of salt-affected soils.

As regards the chemistry of soil salinization and alkalization, two main factors determine the chemical type of salts that dominate in soils and waters. Two factors, climate and landscape geochemistry, are in close correlation and determine whether  $\text{NaCl}$ ,  $\text{Na}_2\text{SO}_4$ ,  $\text{Na}_2\text{CO}_3$  or  $\text{CaSO}_4$ ,  $\text{MgCl}_2$ ,  $\text{MgSO}_4$  will prevail in the processes of salt accumulation.

The parent material, as well as other factors of soil formation, influence the interaction between salts and soils, and it depends on them to a great extent which pedological type will result from such interactions. To mention just one example, solonetz soil will never form in coarse sands, because the development of this type demands a certain amount of highly dispensed soil material.

To understand the regularities of salinization and/or alkalization of a given soil region, and to mitigate or to arrest the harmful processes it is necessary to study the peculiarities of the soil types affected by the accumulation of electrolytes.

#### *Some aspects of salinity and alkalinity in vertisols*

Vertisols are widely distributed in different climatical regions. Some of their properties are similar to that of the solonetz soils. (High clay content, cracking high water retention, etc.) In some places it is difficult even to make clear a distinction between solonetz and vertisol on basis of a superficial land survey.

In many cases, vertisols are affected by salinization and/or alkalization. This process is generally well known, but there are many uncertainties and shortcomings where the influence of electrolytes on vertisols is concerned.

When the accumulation of salts, mainly  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$ , is very intensive and the concentration of them in the topsoil horizons exceeds certain threshold limits, solonchak or saline soils will develop. In such cases, practically independent of the type of original soil material, the effects of the high salt concentration will determine not only the pedological pattern, but also the chemical properties, as well as the agronomical value of the soil.

The picture is not so clear when the concentration of the electrolytes affecting the vertisol is comparatively low. In such cases the sodium ions react to the soil clay material, which is always abundant in vertisols. In spite of the extensive literature describing the rate and character of ion exchange processes in soils, we still lack an exhaustive study of the characteristics of many practical consequences of this phenomenon which differs somewhat in various soils (RICHARDS 1954).

Due to the influence of exchangeable sodium ions, adverse effects will develop in the soil in proportion to the percentage of these exchangeable sodium ions (ESP). It is generally accepted that ESP over 15, which causes formation of columnar B horizon in soils, is associated with the degradation of soil fertility, the decline of hydrophysical properties



and a growing sensitivity to drought. However, it was observed in many countries that the negative effects of exchangeable sodium ions sometimes also appeared when the ESP value was lower than 15; while in other cases, in spite of 20–25 or even higher ESP, soil fertility remained acceptable. Probably under different soil conditions, when the soil type, the mineralogical composition, organic material, etc., are diverse, the threshold values of ESP should be less generalized for all soils, but more associated with the specific soil conditions. Further studies in this direction are important, not only theoretically, but also with a view to the proper evaluation of salt-affected vertisols and other soils, in order to elaborate methods for their utilization and possible amelioration.

As long as the results of basic studies on the impact of salinity and particularly of alkalinity on different soils are not available, local experience should be the guideline for estimating the ESP values, to achieve pedological as well as practical conclusions.

Closely related to the effect of ESP in vertisols, the chemical type of sodium salts and their influence on soil properties should also be studied more concretely in the future.

Most of the available technical literature, particularly the papers discussing irrigation, deal with the problems of salinity induced by NaCl and  $\text{Na}_2\text{SO}_4$ . However, in many places, and often in vertisols, the sodium ions capable of alkaline hydrolysis prevail. The alkaline medium, which often occurs in vertisols, benefits the dominant actions of sodium ions. In such cases, even low sodium concentration may affect soil colloids very intensively, and have a detrimental effect on soil properties and fertility.

To illustrate the specific effects of different sodium salt solutions on soils, in Figs 1 and 2 the results of the experiments, which we conducted on a vertisol from the Hungarian Plain (SZABOLCS 1969) are demonstrated.

The experiments were carried out in soil columns treated with different soil solutions (concentration: 0.1 N). After the treatments we measured the water permeability of soil column.

The data in Fig. 1 show clearly that, while the solutions of NaCl and  $\text{Na}_2\text{SO}_4$  decreased the permeability of soils only moderately, the  $\text{Na}_2\text{CO}_3$  solution made them practically impermeable.

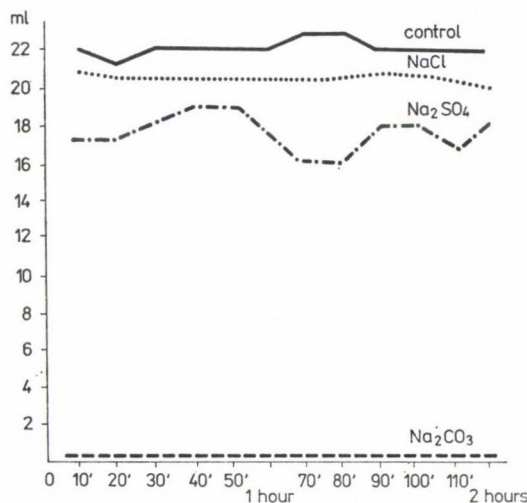


Fig. 1. Water permeability of soils treated with different salt solutions

In Fig. 2 the effect of different sodium salt solutions on soil aggregation is demonstrated. We measured the Vageler-factor in different layers from 0–70–80 cm, in untreated soil as well as in soils, treated with NaCl,  $\text{Na}_2\text{SO}_4$  and  $\text{Na}_2\text{CO}_3$ , respectively.

$$\text{The Vageler-factor} = C = \frac{(a - b) 100}{b},$$

where

$a$  = the amount of the irreversible microaggregates on the basis of the mechanical analysis,  
 $b$  = the amount of the irreversible microaggregates on the basis of the microaggregate analysis.

The results of the experiment, shown in Fig. 2, demonstrate that the destruction of soil aggregates was more intensively affected by  $\text{Na}_2\text{CO}_3$ , than by  $\text{Na}_2\text{SO}_4$  or  $\text{NaCl}$ .

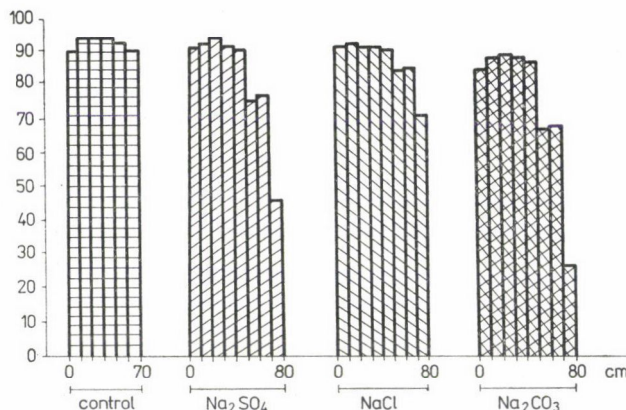


Fig. 2. The structure factor of Vageler in the soils treated with different salt solutions

#### *Main aspects of the assessment of soil salinity and alkalinity*

When assessing soil salinity and/or alkalinity, environmental conditions must be always considered. As salinity and alkalinity occur in different climatical, hydrological and geological conditions, there are no uniform methods to combat those adverse processes. Also the economical conditions of a given area, as well as the patterns of farming, substantially influence the strategies to be elaborated for dealing with salinity and alkalinity.

We must distinguish two main types of assessment:

1. In conditions of rainfed agriculture,
2. In conditions of irrigation.

1. Rainfed agricultural production in salt-affected soils is possible mainly in non-arid conditions. In river valleys and alluvial plains, salinity and alkalinity often occur in semi-humid, but also in humid regions. If the annual precipitation is not sufficient for the removal of excess salts from the soil profile, drainage, and in some cases supplementary irrigation, should be applied in order to gain a yield on salt-affected soils. Under moderate climatic conditions, the yearly distribution of precipitation is often sufficient for keeping the salt balance in soil in such a quasi-equilibrium that the plants can tolerate the salinity and/or alkalinity (International Source Book, 1967).

Where the salt concentration of soils is comparatively low and the main problem is alkalinity causing poor hydrophysical conditions, the solonetz type mainly occur. For the reclamation of alkali soils the application of chemical amendments is a well-known and widely employed method. In non-arid conditions the application of gypsum and other acid products is sometimes effective, even without irrigation. Due to the low degree of salinity in many cases, even moderate doses of gypsum result in the improvement of soils.

Particularly in recent decades, such by-products of industry as phosphogypsum, different  $\text{H}_2\text{SO}_4$  products, iron sulphate, sulphur, etc., have gained popularity in their application, partly due to their availability and low cost.

In some solonetz soils, in the USA, Canada, Hungary, and Argentina, where the horizon A of the soil is slightly acidic, the application of  $\text{CaCO}_3$  and industrial by-products containing such substances is usual.

2. In irrigated areas the problem of salinity and alkalinity has been well known for many thousands of years. The decay of many ancient civilizations was associated with the so-called secondary salinization and alkalization of irrigated soils.

Under arid and semi-arid conditions, irrigation in most cases provokes salinization. It is only in soils which have good natural drainage that irrigation does not cause salt accumulation.

Before constructing irrigation systems in arid conditions, a special survey of soils, hydrological, geological and other environmental conditions should be conducted in order to predict the consequences of irrigation. Potential salinization due to irrigation should be foreseen and, if the hazard is considerable, the areas to be irrigated should be selected in such places that the possibility of adverse processes is less or where they can be controlled.

In Table 2 a scheme of recommended means for the control of secondary salinization and alkalization is demonstrated.

**Table 2**

*Scheme of recommended methods for the control of salinity and alkalinity in irrigated areas*

A. Before construction of irrigation system	Preliminary Survey	
	<i>Landscape</i>	<i>Planned irrigation</i>
	climate	available irrigation water (quantity and quality)
	hydrology	
	hydrogeology	ground water (depth and quality)
	geomorphology	technology of irrigation cropping pattern (tolerance)
B. During irrigation	Monitoring:	salinity and alkalinity of soil
		groundwater table
		chem. comp. of groundwater
		chem. comp. or irrigation water filtration
		physical soil properties
		possible toxic elements in soil and water (B, etc.)

Table 2 shows that the prediction of secondary salinization and alkalization of the soils to be irrigated should be based on a preliminary survey of the landscape and soils before the construction of irrigation systems. Thus, it is possible to take the necessary steps to prevent these adverse processes.

During the irrigation, a well organized monitoring of soil and water properties is necessary in order to note the changes, if any, and to take necessary precautions. The methods for monitoring, the timing and places of sampling depend on local conditions.

### SUMMARY

Soil salinity and alkalinity may develop in different areas and are associated with many soil-forming processes. Depending on soil types and local environmental conditions, salinity and alkalinity influence the soil and the landscape diversely.

Arid conditions contribute to salinization, but alkalization particularly prevails also in semi-arid and semi-humid areas.

The threshold values of ESP which cause poor hydrophysical properties in soils have not been exactly elaborated. In many cases, such as in vertisols, anomalies have been observed in the application of 15–20 ESP values as characteristics for solonchaks. Soil research aiming at a more precise understanding of the interaction of cations with soil colloids should be done, giving more consideration to the different soil types.

The management of salinity and alkalinity of soils differs in rainfed and in irrigated conditions. Secondary salinization and alkalization are old but still-growing hazards in most irrigated areas. Preventive means and permanent monitoring are necessary to control these processes.

\*



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### References

- RICHARDS, L. A. (ed.) (1954): Diagnosis and improvement of saline and alkali soils. Agric. Handbook No. 60. USDA, Washington.
- International Source-Book on irrigation and drainage of arid lands in relation to salinity and alkalinity. 1967. FAO-UNESCO, Paris.
- KOVDA, V. A. (1946-47): Origin and regime of salt-affected soils. I—II. (In Russ.) Moscow, Acad. Sci. USSR.
- POLYNOV, B. B. (1956): Selected papers. (In Russ.) Acad. Sci. USSR, Moscow.
- LYNN, W. C. and MCKINZIE, W. E. (ed.) (1976): Soil Taxonomy. USDA, Lincoln.
- Soil Map of the World 1 : 5 000 000 FAO-UNESCO, Paris.
- SZABOLCS, I. (1969): The influence of sodium carbonate on soil-forming processes and on soil properties. *Agrokémia és Talajtan*, **18**, Suppl. 37-68.
- SZABOLCS, I. (1981): Landscape geochemistry of soil salinization and alkalization. *Agrokémia és Talajtan*, **30**, Suppl. 47-62.

### CHANGES IN THE TRAITS OF MAIZE HYBRIDS DEVELOPED FROM MUTANTS\*

With the general use of hybrid varieties of maize, the basis for the foundation material of maize improvement has significantly decreased. It is a further problem for some countries (including Hungary) that the most generally grown hybrids are to a certain degree related to each other. This may be dangerous as it decreases the stability of maize yields. Therefore the foundation material used for breeding should be increased and the gene pool should be enriched, in which task radiomutational research can greatly assist.

Mutational research can help much in developing new varieties, better suited to the requirements of production and utilization (e.g. early maturing, high yielding, disease resistant, standable types or types with erect leaves, more ears, or high in protein, macro- and microelement content, etc.). They open up new vistas to satisfying the variety needs of plant production and to developing hybrids of better genetic structure.

Improvement by mutation differs to a certain degree from traditional methods of plant improvement. It takes more time but it offers better prospects in the long run than do traditional methods. So far we have selected about 250 mutant lines for various morphological traits, the length of the vegetation period, etc. and we are maintaining them by inbreeding.

In the course of our experimenting so far with mutants, we have observed that some traits become fixed in them. We got some hybrids with favourable traits by crossing these mutants with normal lines. Some other mutants, however, kept developing various traits in spite of long periods of self-pollination. Our experiments show that there is a constant variety-forming process at work in the mutant population, while — owing to inbreeding — some characteristics that occur are easily fixed.

In the time now available I cannot analyse the results of our research in detail. But a few data may indicate the possibilities of radiomutational maize breeding to increase the yields and improve the quality traits of maize.

We got the following results at the experimental farm of the Agricultural University of Debrecen, and the data have been supplied by the National Institute for Agricultural Variety Experiments — OMFI — of Hungary.

We conducted our experiments with grain maize on areas of 70×30 cm and 70×25 cm (47 617 plants/ha and 57 143 plants/ha, respectively). With the exception of the standard lines, one of the parent lines of the hybrids was a mutant. Let me present now some data pertaining to the results of experiments conducted both with grain maize and with silage maize.

\* Lecture held at the XII. meeting of the ESNA (European Society of Nuclear Methods in Agriculture) in Aberdeen Scotland from 28th September to 2nd October, 1981.

### *The grain yield of hybrids*

In the experiments conducted in 1977, DSC 2584 had the highest grain yield (Table 1).

It yielded 10% more grain than the st. (0.95 t/ha) and its protein yield was also higher by 0.13 t/ha. In the other two hybrids, protein yield could be increased by 0.14 and 0.46 t/ha, respectively, but their grain yields (compared to the st.) decreased by 23% and 7%, respectively. This goes to show that it is not always possible to increase grain yield and protein content simultaneously. These hybrids were analysed for amino acid content, too. DSC 2584 excelled in methionine and lysine content.

In 1978, hybrids originating from crossing various mutant and normal lines were examined for grain yield and protein content. As Table 1 shows, by using these various mutant



*Fig. 1. DSC-2584 and its parents*

lines, grain yield and protein content could be simultaneously improved. E.g. in K-3563  $\times$  M-55, GK-2  $\times$  M-123, K-3563  $\times$  M-13 and 156  $\times$  M-26. There are some hybrids whose grain yields are the same as, or lower than, that of the standard; but their protein yield per ha is higher, because their protein content is higher.

Our experiments in 1979 with four-line hybrids gave similar results. As the data of Fig. 1 show, grain yield and protein content could be simultaneously increased in these hybrids (J-59  $\times$  RDSB-1431)  $\times$  (GK-2  $\times$  M-123), (J-59  $\times$  Sz-2351)  $\times$  (GK-2  $\times$  M-141).

### *Hybrids examined for silage value*

Table 3 shows the results of experiments conducted in 1979 and 1980 with hybrids for silage use. One of the parents of these hybrids, too, was a mutant. The results of the experiments clearly show that mutants can be used for breeding maize for silage use as well. Some tillering mutant lines are especially suitable for this purpose. The data in the Table 3 illustrate that the ratio of ears could be increased simultaneously with increasing the yield of green and dry matter (ZMK-3  $\times$  B-14, SC-2585, N-6  $\times$  M-10).

One of the parents of ZMK-3 was an Opaque-2 type mutant. One of the parents of SC 2585 was a J-59 line, which is again a mutant. The mutant lines in the other hybrids were

**Table 1**  
*The grain yields and protein contents of two- and four-line hybrids*  
 (Debrecen, 1977-1979)

Crossing combinations	Grain yield, t/ha	%	Crude protein, %	Protein yield, t/ha
<i>1977</i>				
1. MvSC-580 st.	9.95	100	10.66	1.06
2. DSC-2584	10.90	110	10.88	1.19
3. DSC-344	7.72	77	14.88	1.20
4. DSC-415	9.29	93	16.38	1.52
LSD <sub>5%</sub>	0.85			
<i>1978</i>				
1. MvSC-580 st.	11.11	100	10.13	1.13
2. K-3563 × M-55	13.15	118	12.13	1.60
3. K-3563 × M-114	13.44	121	8.23	1.11
4. K-3563 × M-120	14.89	134	9.56	1.42
5. K-3563 × M-48	15.67	141	9.96	1.56
LSD <sub>5%</sub>	2.77			
1. MvSC-580 st.	15.41	100	10.92	1.69
2. GK-2 × M-119	14.19	92	12.35	1.75
3. GK-2 × M-71	15.06	98	11.68	1.76
4. GK-2 × M-137	15.22	99	11.46	1.74
5. GK-2 × M-123	15.50	101	11.91	1.85
LSD <sub>5%</sub>	4.46			
1. MvSC-580 st.	13.17	100	11.25	1.48
2. 156 × M-28	12.51	95	11.34	1.42
3. 156 × M-119	13.35	101	9.69	1.29
4. K-3563 × M-13	14.55	111	10.25	1.49
5. 156 × M-26	15.96	121	11.59	1.85
LSD <sub>5%</sub>	2.12			
<i>1979</i>				
1. MvDC-460 st.	13.34	100	11.90	1.59
2. (J-59 × RDSB-1431) × (GK-2 × M-123)	14.24	107	11.41	1.62
3. (J-59 × Sz-2351) × (156 × M-11)	16.23	122	9.75	1.58
4. (J-59 × Sz-2351) × (GK-2 × M-141)	16.94	127	12.13	2.05
LSD <sub>5%</sub>	3.35			



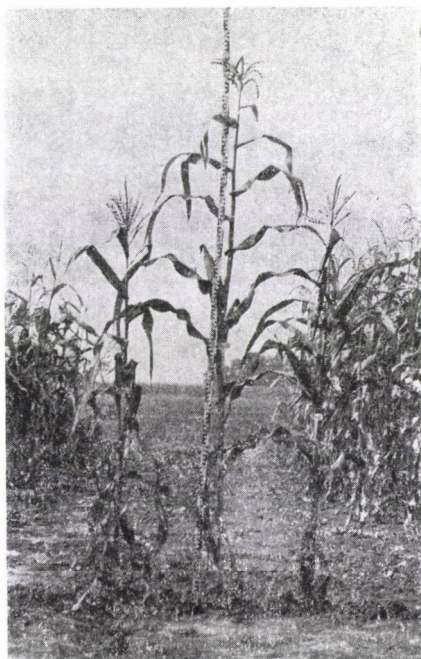


Fig. 2. SC-2390 and its parents

Table 2

*The aminoacid contents of various hybrids*  
(Debrecen, 1977)

Aminoacids	Hybrids		
	DSC 2584	DSC 344 (K-3562 × J-59)	DSC 415 (PTF-3943 × J-59)
Crude protein	10.88	14.88	16.38
	g/16 g N/100 protein		
Asparatic acid	7.36	6.37	6.93
Threonine	2.99	2.68	2.18
Serine	3.56	3.10	2.96
Glutamin acid	25.73	28.35	24.42
Proline	6.32	5.87	6.70
Glycine	3.78	3.61	3.42
Alanine	8.27	7.13	5.63
Cystine	0.83	0.87	0.92
Valine	5.06	4.45	5.63
Methionine	2.47	2.33	2.19
Isoleucine	3.68	4.19	3.58
Leucine	14.34	14.60	13.93
Tyrosine	2.41	2.18	2.73
Phenyl alanine	5.06	4.53	4.10
Lysine	2.18	1.43	2.23
Arginine	2.53	2.35	2.44
Tryptophan	0.68	0.40	0.38

**Table 3**  
*The green matter, dry matter and protein yield of various hybrids*  
 (Debrecen, 1979–1980)

Crossing combinations	Green matter yield (stem+leaf+ ear), t/ha	Dry matter yield, t/ha	Ear-dry matter, %	Total protein yield, t/ha
<i>1979</i>				
1. MvDC 460 st.	51.8	14.6	50.7	1.17
2. SC-2584	59.3	20.2	40.6	1.47
3. SC-2585	56.7	20.8	47.1	2.08
4. (K-3563 × ZMK-3) × J-59	48.8	16.8	53.0	1.51
5. SC-2390	46.7	14.9	59.1	1.38
6. ZMK-3 × B-14	57.4	17.5	49.1	1.47
LSD <sub>5%</sub>	6.98			
<i>1980</i>				
1. SzaDC-590 st.	36.8	10.3	45.3	0.76
2. SC-2390	39.1	14.8	48.6	0.88
3. (K-3563 × ZMK-3) × J-59	33.1	11.1	44.4	0.74
4. T-27 × RDSB-3712	38.3	10.8	44.5	0.94
LSD <sub>5%</sub>	14.3			
1. SzaDC-590 st.	53.3	15.0	45.4	1.12
2. C-5 × M-81	40.8	12.4	45.5	1.01
3. N-6 × M-10	55.4	16.1	45.7	1.03
LSD <sub>5%</sub>	9.99			

indicated by an M. SC-2390 originated from a cross between the ZMK-3 and the J-59 lines and was entered for the national variety experiments in 1978.

SC-2390 for silage use did well at the national experiments of 1979 and 1980.

The green matter yield of SC-2390 was 49.85 t/ha in the two years' average. It yielded 2.1 t/ha more than the st., whose yield was 47.7 t/ha. The Q-2 silage hybrid yielded higher in green matter than the two standards in both years (Table 4). Its dry matter yield was 15.62 t/ha, which was 0.63 t/ha more than the yield of the standard (which was 14.99 t/ha in the two years' average). The protein yield of this Q-2 type silage hybrid (SC-2390) was 1.24 t/ha, while that of the standard was 1.07 t/ha. This means 170 kg more protein per ha, which is 15.3% higher than the protein yield of the standard.

The absolute value of the protein yield of this silage maize is not low, either. It is not only for its green and dry matter yield that the Q-2 type silage hybrid is remarkable but mainly for its protein yield per ha. Its total carbohydrate yield, too, was higher than that of the standard. The results of resistance tests were also good.

Another hybrid of ours, SC-2584 did similarly well on the national silage maize experiments after it had shown good results as grain maize, too, in previous years.

Our SC-2584 silage hybrid had excellent results on the silage experiments of 1980. It yielded the highest amounts of dry matter and protein in these experiments. Compared with the standard, its dry matter yield was 40% higher and its crude protein yield was 46.4% higher (Table 5). This hybrid took first place in 1980 for its high protein yield.

### Conclusion

By presenting the results of our experiments, we aimed to draw your attention to the applicability of mutants to maize improvement. It deserves attention that the hybrids presented here excel especially in their higher yields of protein (t/ha). By using the right kind of hybrids, by increasing grain yield, protein yield can be increased, too. Choosing the right hybrids is important because there are some whose grain yields decrease although their protein yields increase. By using mutant lines, we managed to develop silage hybrids with not only increased yields of protein, dry and green matter, but also with an improved ear-

### Table 4

*The results of examinations done into the quality traits of SC-2390 hybrid maize and st. in 1979-80*

(National experiments done by the OMFI)

Stem + leaf + ear	SC 2390	st.	% compared to st.	Deviation from the st. $\pm$
<i>1979 (7 experiments), t/ha</i>				
Green matter yield	54.01	50.65	106.6	+3.36
Dry matter yield	16.66	15.82	105.3	+0.84
<i>Total protein yield</i>	<i>1.31</i>	1.09	<i>120.2</i>	+0.22
Total crude fibre	3.75	3.92	95.6	-0.23
Total carbohydrate (sugar + starch)	10.35	9.27	116.6	+1.08
<i>1980 (6 experiments), t/ha</i>				
Green matter yield	45.70	44.80	102.0	+0.90
Dry matter yield	14.59	14.17	102.9	+0.42
<i>Total protein yield</i>	<i>1.17</i>	1.06	<i>110.4</i>	+0.11
Total crude fibre	3.36	3.15	106.6	+0.21
Total carbohydrate (sugar + starch)	8.69	8.64	100.5	+0.05
<i>The average of the two years</i>				
Green matter yield	49.85	47.72	104.4	+2.13
Dry matter yield	15.62	14.99	104.2	+0.63
<i>Total protein yield</i>	<i>1.24</i>	1.07	<i>115.3</i>	+0.17
Total crude fibre	3.55	3.53	100.5	+0.02
Total carbohydrate	9.52	8.95	106.1	+0.57

Notes: Cr. protein:

max.

min.

1979 1.31 (SC-2390) 1.09 (M<sub>v</sub>-26)

1980 1.55 (SC-2584) 1.06 (Mv-26)

St. 1979 (SzDc-590), 1980 (Mv-26, SzDc-590)

Table 5

*The results of examinations into the quality traits of SC-2584 hybrid maize and st. in 1980*

(National experiments by the OMFI)

Stem + leaf + ear	SC 2584	Mv-26 st.	Hybrid-st. %	LSD 5%
<i>1980 (5 experiments)</i>				
1. Green matter yield, t/ha	49.79	44.80	111.1	4.60
2. Dry matter yield, t/ha	19.86	14.17	140.2	1.78
3. Ear-dry matter, %	51.20	37.90	135.4	12.64
4. Total protein yield, t/ha	1.55	1.66	146.4	
5. Total crude fibre, t/ha	3.93	3.15	124.5	
6. Total N-free extract	12.60	8.65	145.9	



ratio. Their macro- and microelement content was better, too. These hybrids surpass the standards in several traits in the course of their phenomenological development.

All our experiments conducted so far have proved that mutants can be used in breeding mainly to improve the quality of maize.

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## A STUDY OF THE AGRONOMICAL AND CHEMICAL CHARACTERISTICS OF MAIZE HYBRIDS SC-2390 AND SC-2584 PRODUCED FROM MUTANT LINES

The main areas of further development in maize production are yield safety, chemical composition and better utilization (KOVÁCS and PÁSZTOR 1981). Mutant populations of richer varieties provide considerable help in solving these problems (BÁLINT 1967, 1977, PÁSZTOR 1978, 1980).

We have continued our research of the previous years into the selection of maize mutants as well as our studies of the combining abilities and chemical characteristics of hybrids. The aim of these experiments is to determine the improvement values of mutant lines both for chemical characters and for yield components. The development of animal husbandry and maize production necessitated improving the chemical composition of maize. The animal organism can only function normally when provided with certain indispensable microelements like Fe, J, Cu, Mn, Co in its food, as these elements take part in the vitally important processes of metabolism and synthesis (DINGA and TABARU 1970). Microelements influence the activity of various enzymes, control protein- and lipogenesis as well as metabolism, growth, reproduction and milk yield (BERGMANN and HENNIG 1973). Deficiencies in phosphorus, potassium, magnesium and cobalt have a detrimental effect on animal health (STEWART 1965). The development of maize-processing industries (the production of oil, starch, liquid sugar, distilling, etc.) sets up further requirements. When improving maize, its various uses should be taken into account, not only considering the quantity of yield but also its composition (SÁGI 1979).

This paper contains but a small part of the research carried out in this field.

Experiments were conducted at the Experimental Station of the Agricultural University of Debrecen and by the National Agricultural Variety Testing Institute. The experiment in Debrecen was carried out on chernozem soil, incrustated with lime. The depth of the humus layer varied between 40–80 cm, and the depth of water in the subsoil between 8–12 m. The pH value was 7.3; the humus content 3.5–3.7%. The Arany viscosity index was 34, and the forecrop was peas. The area was fertilized by 120 kg N, 83 kg  $P_2O_5$  and 100 kg  $K_2O$ .

The hybrids tested in the experiment were two- or three-line ones with one of the parents being a mutant line. The maternal line of hybrid SC-2390 was an Opaque mutant (MQ<sub>2</sub>-1). Its paternal line (DPM-2-59) was selected from an inbred line that had been irradiated during the vegetation period by a dose of 7 Gy (of a  $^{60}Co$  radiation source) in the gamma-field of the Agricultural University of Gödöllő. "M" marks parent lines selected from mutant populations, that were produced in 1958 from local strains by treating their pollens with a dose of 15 Gy (of  $^{60}Co$ ). The above two hybrids (SC-2390 and SC-2584) have been tested in national variety comparison experiments for years.

The hybrids and their parent lines underwent various tests (phenological ones, determination of yield components, chemical characteristics, etc.). N content was determined by the macro-Kjeldahl method, while P content was determined by the molybdovanadate method. K content was determined by a flame photometer, while the contents of Mg, Mn, Zn and Cu were determined by an atom absorption spectrophotometer.

The results were mathematically evaluated by variance analysis.

The results of small plot experiments (started in 1980) with hybrids SC-2390 and SC-2584, as well as their parent lines, were analysed in detail. The performance of the two hybrids in the national variety tests, as well as the results of the three-line (TC) variety of the SC-2390 hybrid, were evaluated.

\* Lecture held at the meeting of the ESNA (European Society of Nuclear Methods in Agriculture) in Brno, CSSR, from 6th September to 11th September, 1982.

*Hybrid SC-2390 and its parent lines*

Tables 1 and 2 show the data of experiments conducted in 1980 with hybrid SC-2390 and its parent lines.

As can be seen from the data of Table 1, when compared with the parent lines, the hybrid showed a considerable heterosis effect in plant height, number of leaves, surface area of leaves, length of tassel and number of lateral branches of the tassel. Of the examined characteristics, side-shoot producing percentage had a high CV value (Table 5).

When the chemical characteristics of the parent lines were compared with those of the hybrid SC-2390, the heterosis effect was pronounced in the K and Zn contents of the grain; the protein, Mg, Cu and Mn contents of the stem, and the K, Zn and Mn contents of the leaves.

*Hybrid SC-2584 and its parent lines*

The results of experiments made in 1980 with hybrid SC-2584 and its parent lines are summed up in Tables 3 and 4. The CV values of the two hybrids (SC-2390 and SC-2584) and of their parent lines can be found in Table 5.

The data presented in Table 3 illustrate the considerable heterosis effect in hybrid SC-2584 in plant height, thickness of the stem, length of leaves, surface area of leaves, side-shoot producing percentage, length of the tassel, number of lateral branches of the tassel, number of ears per plant and the ratio of ears per plant parts.

Table 4 shows, from the chemical characteristics, the Cu and starch contents of the grains; the K, Ca and Cu contents of the stem; the P, K, Zn and Mn contents of the leaves of the hybrids are higher; while the protein, P, K, Ca, Mg and Mn contents of the grain, the K, Mg and Mn contents of the stem, and the protein, K, Mg and Mn contents of the leaves, are lower than those of the parent plants.

Side-shoot producing percentage varies rather widely (the CV value is high).

**Table 1**

*Phenologic characteristics of the hybrids SC-2390 and of their inbred lines  
(Debrecen, 1980)*

Characteristics	Parents (P)	Hybrids (F <sub>1</sub> )		Average $\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$
	MQ <sub>2</sub> -1	DPM-2-59	SC-2390			
1. Plant height, cm	214.3	157.8	237.5	186.1	51.4	0.276
2. Stem thickness, mm	27.6	25.8	29.4	26.7	2.7	0.101
3. Stem thickness (Plant height %)	1.288	1.635	1.238	1.462	-0.224	-0.153
4. Number of leaves	15.1	13.8	17.6	14.5	3.1	0.214
5. Length of leaves, cm	80.6	71.2	87.6	75.9	11.7	0.154
6. Breadth of leaves, cm	11.3	11.5	12.2	11.4	0.8	0.070
7. Leaf surface area, cm <sup>2</sup>	683.1	614.1	801.5	648.6	152.9	0.236
8. Number of internodes	13.2	12.1	13.5	12.7	0.8	0.063
9. Side-shoot producing (%)	5.3	7.1	4.8	6.2	-1.4	-0.226
10. Length of tassel, cm	34.7	31.9	40.5	33.3	7.2	0.216
11. Number of lateral branches of tassel	19.6	17.5	24.9	18.6	6.3	0.339
12. Number of ears per plant	1.11	1.43	1.87	1.27	0.6	0.472
13. Plant proportions						
— ear %	32.62	34.38	37.03	33.50	3.53	0.105
— stem %	34.67	35.15	35.06	34.91	0.15	0.004
— leaf %	32.71	30.47	27.91	31.59	-3.68	-0.117

Table 2

*Heterosis in the chemical composition of  
(Debrecen,*

Chemical characters	Grains						Stem		
	MQ <sub>2</sub> -1	DPM 2-59	SC-2390	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	MQ <sub>2</sub> -1	DPM 2-59	SC-2390
Protein, %	10.56	10.88	9.13	10.72	— 1.59	—0.148	5.38	3.00	6.81
P, %	0.242	0.268	0.260	0.255	0.005	0.020	0.178	0.135	0.105
K, %	0.290	0.260	0.330	0.275	0.055	0.200	1.73	2.06	1.544
Ca (ppm)	93.0	114.0	77.0	103.5	—26.5	—0.256	1750.0	2000.0	1650.0
Mg (ppm)	960.0	1000.0	980.0	980.0	∅	∅	760.0	560.0	800.0
Zn (ppm)	11.8	17.0	17.40	14.4	3.0	0.208	18.0	27.0	22.0
Cu (ppm)	0.70	0.90	1.45	0.80	0.65	0.813	3.6	5.0	6.5
Mn (ppm)	5.2	6.4	4.8	5.8	— 1.0	—0.172	10.0	9.00	12.0
Starch, %	56.33	52.54	56.33	54.44	1.89	0.035	—	—	—
Fiber, %	—	—	—	—	—	—	31.24	25.95	28.18

Table 3

*Phenologic characteristics of the hybrids SC-2584 and of their inbred lines  
(Debrecen, 1980)*

Characteristics	Parents (P)		Hybrids (F <sub>1</sub> )	Average P	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$
	DPK 1-59	B-14	SC-2584			
1. Plant height, cm	146.7	180.5	245.8	163.6	82.2	0.502
2. Stem thickness, mm	22.3	31.1	32.8	26.7	6.1	0.229
3. Stem thickness (plant height %)	1.520	1.723	1.334	1.622	—0.288	—0.178
4. Number of leaves	13.8	16.1	17.5	15.0	2.5	0.167
5. Length of leaves, cm	69.9	86.4	103.2	78.2	25.0	0.320
6. Breadth of leaves, cm	9.2	10.2	10.7	9.7	1.0	0.103
7. Leaf surface area, cm <sup>2</sup>	482.3	661.0	828.2	571.7	256.5	0.449
8. Number of internodes	11.6	14.3	15.0	13.0	2.0	0.154
9. Side-shoot producing, %	15.52	0.05	20.00	7.79	12.21	0.611
10. Length of tassel, cm	30.8	36.3	45.6	33.6	12.0	0.357
11. Number of lateral branches of tassel	12.3	7.7	16.4	10.0	6.4	0.640
12. Number of ears per plant	1.47	1.14	1.85	1.31	0.54	0.412
13. Plant proportions						
— ear, %	16.62	23.15	31.77	19.89	11.88	0.597
— stem, %	46.76	46.50	43.91	46.63	—2.72	—0.058
— leaf, %	36.62	30.35	24.32	33.49	—9.17	—0.274



hybrids SC-2390 and the inbred lines  
1980)

Stem			Leaves					
$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	$MQ_{r-1}$	DPM 2-59	SC-2390	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$
4.19	2.62	0.625	7.69	11.06	6.63	9.38	— 2.75	— 0.293
0.157	— 0.052	— 0.331	0.180	0.180	0.160	0.180	— 0.020	— 0.111
1.90	— 0.356	— 0.187	1.02	0.70	1.28	0.86	0.42	0.488
1875.0	— 225.0	— 0.120	6700	4900	4900	5800	— 900	— 0.155
660.0	140.0	0.212	1760	1180	1120	1470	— 350	— 0.238
22.5	— 0.5	— 0.022	12.5	12.5	15.0	12.5	2.5	0.200
4.3	2.2	0.512	2.7	2.1	2.1	2.4	— 0.3	— 0.125
9.5	2.5	0.263	40.5	33.0	62.0	36.8	25.2	0.685
—	—	—	—	—	—	—	—	—
28.60	— 0.42	— 0.015	25.86	27.26	28.55	26.60	1.95	0.073

*The performance of hybrids SC-2390 and SC-2584 in the National Variety Comparison Tests*

Of the two hybrids, SC-2390 was entered for the variety certification tests in 1978, and SC-2584 in 1980. The results of the national small-plot experiments of the National Variety Testing Institute (Table 6) show that hybrid SC-2390 surpassed the standard in the average of two years by 4% in dry matter yield, by 15% in total protein yield, by 10% in crude fiber content, and by 6% in total N-free extractable matter.

In 1981 hybrid SC-2390 surpassed the standard (Sze DC-538) in dry matter yield by 3.9% (19.8 t/ha and 19.0 t/ha, respectively). The green matter yield of the hybrid was 46.4 t/ha, which as 6.4% less than that of the standard. The hybrid's total protein yield was 1.57 t/ha, its total crude fiber yield was 4.66 t/ha, and its total N-free extractable matter yield was 11.80 t/ha. In comparison, the protein yield of the standard (Sze DC-538) was 1.60 t/ha, its total crude fiber yield was 4.10 t/ha, while its total N-free extractable matter yield was 11.71 t/ha.

The results of the 1980 experiment conducted by the NVTI (Table 6) show that hybrid SC-2584 surpassed standard Mv26 by 11% in green matter yield, by 40% in dry matter yield, by 46% in total protein yield, by 25% in crude protein yield and by 46% in total N-free extractable matter yield.

In the 1981 national experiment, hybrid SC-2584 had a dry matter yield of 18.96 t/ha, a green matter yield of 45.29 t/ha, a protein yield of 1.55 t/ha, a total crude fiber yield of 4.46 t/ha and a total N-free extractable matter yield of 11.27 t/ha. This year its performance has reached the level of standard Sze DC-538. In 1981 the value of LSD 5% was 2.5 t/ha in dry matter yield and 5.06 t/ha in green matter yield for both hybrids. Our hybrids surpass or equal the performance of most experimental varieties this year, too, nor do they fall significantly behind any of the other varieties. This tendency is even more marked in the average of several years.

*TC hybrids*

In 1980 hybrid SC-2390 was used as the maternal line in crossings with two mutant lines of different genotypes. The three-line (TC) hybrids thus developed were studied in small-plot comparison experiments at the Experimental Station of the Agricultural University of Debrecen in 1981. The results can be seen in Table 7.

Table 4

*Heterosis in the chemical composition of  
(Debrecen,*

Chemical characters	Grains						Stem		
	DPK 1-59	B-14	SC-2584	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	DPK 1-59	B-14	SC-2584
Protein, %	9.00	11.75	9.19	10.38	— 1.19	—0.115	11.94	7.44	6.38
P, %	0.3	0.32	0.294	0.31	— 0.016	—0.05	0.27	0.15	0.24
K, %	0.4	0.47	0.32	0.44	— 0.12	—0.27	0.74	1.42	0.57
Ca (ppm)	151.0	98.0	77.0	124.5	—47.5	—0.38	1800.0	1650.0	2400.0
Mg (ppm)	980.0	1090.0	960.0	1035.0	—75.0	—0.07	1520.0	1160.0	1020.0
Zn (ppm)	11.8	14.2	14.6	13.0	1.6	0.12	15.0	25.0	21.0
Cu (ppm)	0.90	1.45	2.08	1.18	1.62	1.37	3.4	3.4	7.6
Mn (ppm)	6.8	6.0	4.8	6.4	— 1.6	—0.25	19.0	78.0	7.0
Starch, %	38.46	38.46	49.83	38.46	11.37	0.30	—	—	—
Fiber, %	—	—	—	—	—	—	24.15	30.46	26.88

It is evident from the data of Table 7 that, by crossing two-line hybrid SC-2390 with mutant line M-122, dry matter yield can be increased considerably (24 323 kg/ha). When the same two-line hybrid was crossed with mutant line M-197, the increase was even greater (29 314 kg/ha). The hybrid surpassed both SC-2390 and the two Pioneer hybrids in green and dry matter yield, and the protein content of the plant parts was also high (Table 7).

Table 5

*Value of the relative standard deviation (CV %) of improved hybrids and inbred lines  
(Debrecen, 1980)*

Characteristics	Parent (P)		Hybrids ( $F_1$ )	Parents (P)		Hybrids ( $F_1$ )
	DPK 1-59	B-14	SC-2584	MQ <sub>2</sub> -1	DPM 2-59	SC-2390
1. Plant height, cm	3.64	4.29	6.28	5.12	4.15	5.94
2. Stem thickness, mm	7.35	8.10	9.23	7.99	6.57	9.14
3. Number of leaves	2.75	3.62	5.42	4.23	3.67	6.58
4. Length of leaves	5.73	5.51	6.78	5.79	4.27	6.05
5. Breadth of leaves, cm <sup>2</sup>	8.27	7.38	9.85	7.38	7.59	8.76
6. Number of internodes	2.28	3.05	4.79	4.25	4.64	4.39
7. Side-shoot producing, %	17.34	5.26	19.78	14.23	18.24	15.67
8. Length of tassel	5.73	6.27	8.14	7.15	5.43	8.65
9. Number of lateral branches of tassel	8.25	7.38	9.39	7.26	6.53	8.10
10. Number of ears per plant	4.12	3.24	5.18	3.28	5.27	6.15
11. Plant proportions						
— ear, %	6.23	7.42	8.56	6.14	6.28	7.56
— stem, %	7.42	7.93	8.14	6.25	6.32	6.47
— leaf, %	7.12	6.48	7.65	8.25	7.63	6.15

hybrids SC-2584 and the inbred lines  
1980)

Stem				Leaves					
$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	DPK 1-59	B-14	SC-2584	$\bar{P}$	$\bar{F}_1 - \bar{P}$	$\frac{\bar{F}_1 - \bar{P}}{\bar{P}}$	
9.69	— 3.31	—0.342	9.38	11.75	8.50	10.57	— 2.07	—0.196	
0.21	0.03	0.143	0.22	0.22	0.3	0.22	0.08	0.364	
1.08	— 0.51	0.472	0.56	0.5	0.96	0.53	0.43	0.811	
1725.0	675.0	0.391	5600.0	5300.0	5300.0	5450.0	150.0	—0.028	
1340.0	—320.0	—0.239	1720.0	2660.0	1760.0	2190.0	—430.0	—0.196	
20.0	1.0	0.050	8.0	15.0	15.0	11.5	3.5	0.304	
3.4	4.2	1.235	2.1	4.2	3.4	3.2	0.2	0.063	
48.5	— 41.5	—0.856	48.8	11.0	55.0	29.9	25.1	0.840	
—	—	—	—	—	—	—	—	—	
27.31	— 0.43	—0.016	26.46	24.75	24.55	25.61	— 1.06	—0.04	

Summing up the results of the experiments, it can be ascertained that lines originating from crosses with mutants can be of great help in producing hybrids of superior genetical structure.

— There is a considerable heterosis effect in certain phenological and morphological characters (plant height, number of leaves, surface area of leaves, tassels, etc.).

— The averages of green and dry matter yields of several years of the two hybrids surpass those of the standards.

Their chemical composition is favourable, too; protein yield being especially high.

The dry matter yield of various three-line (TC) hybrids — where different mutant lines were used as paternal lines — increased considerably (24.32 and 29.31 t/ha), and protein yields were also quite good.

\*

Prepared at the Exp. Station of the Agricultural Univ. of Debrecen and Mosonmagyaróvár by the National Agricultural Variety Testing Institute, Hungary

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### References

- BÁLINT, A. (1967): Heterosis and Mutation in Maize. Akadémiai Kiadó, Budapest.  
 BÁLINT, A. (1977): The Production Genetics of Farm Plants. Akadémiai Kiadó, Budapest.  
 BERGMANN, W. and HENNIG, A. (1973): Bedeutung der Spurelemente im Rahmen der weiteren Intensivierung der landwirtschaftlichen Produktion. Chem. Techn., **25**, 7.  
 DINGA, D. and TABARANU, T. (1970): Influența ingrasamintelor asupra continutului in microelemente (Mn, B, Cu) din principale plante de nutreț din zona solului brun roscat de padure. Anale Institutului de Cercetari Pentru Cereale și Plante Technice, Fundulea. Seria B. București, **36**, 639.  
 PÁSZTOR, K. (1978): Effect of Mutagens on the Variability of Morphological Characters in Maize. Acta Agronomica Academica Scientiarum Hungaricae. **22**, 3–4, 481–488.  
 PÁSZTOR, K., HASSAN, R. and ELEK, M. (1980): An Evaluation of the Chemical Composition of Maize Mutants and of their Hybrids. Növénytermelés, **29**, 1, 9–20.  
 SÁGI, F. (1979): The Quality of Maize. Budapest, Agroinform, 86.  
 STEWART, A. B. (1965): Aspects of Soil, Plant and Animal Relationships. Fertiliser and Feeding Stuffs Journal, London, **62**: **18**, 709–717.



Table 6

*The chemical composition of hybrids SC-2390 and SC-2584 and a standard silage maize (INVTI 1979–1980)*

Leaves + stem + ears, t/ha	SC-2390						SC-2584				
	1979			1980			The average of 2 years			1980	
	Hybrid	SDC-590 st.	Hybrid/ st. %	Hybrid	Mv. 26 st.	Hybrid/ st. %	Hybrid	st.	Hybrid/ st. %	Hybrid	Hybrid (Mv. 26 st.) %
Green matter yield	54.01	50.65	106.6	45.70	44.80	102.0	49.85	47.72	104.4	49.79	111.1
Dry matter yield	16.66	15.82	105.3	14.59	14.17	102.9	15.62	14.99	104.2	19.86	140.2
Total protein yield	1.31	1.09	120.2	1.17	1.06	110.4	1.24	1.07	115.3	1.55	146.4
Total crude protein yield	3.75	3.92	95.6	3.36	3.15	106.6	3.55	3.53	110.5	3.93	124.5
Total N-free extract.	10.35	9.27	116.6	8.69	8.64	100.5	9.52	8.95	106.1	12.60	145.7

1979 SD<sub>5%</sub> for green matter yield 3.55 t/ha, SD 5% for dry matter yield 1.49 t/ha  
 1980 SD<sub>5%</sub> for green matter yield 1.78 t/ha, SD 5% for dry matter yield 4.60 t/ha

Table 7

*The green and dry matter yields, protein contents and fiber percentages of hybrids of various genotypes (Debrecen, 1980)*

Inbred lines and hybrids	Green matter yield	Dry matter yield	Protein, %				Fiber, %			
			ears	stem	leaves	husks	ears	stem	leaves	husks
1. MQ <sup>2</sup> -1	60.713	16.984	8.75	5.63	10.00	6.25	8.60	32.67	27.06	28.40
2. SC-2390	73.429	21.440	9.385	3.75	8.75	5.00	10.76	32.25	27.92	37.06
3. DP-M-2-59	52.286	16.159	11.86	6.25	8.13	7.50	6.87	32.63	26.14	28.60
4. SC-2390 × M-122 (TC)	72.000	24.323	9.38	4.38	4.38	9.38	5.68	36.46	35.41	29.14
5. M-122	—	—	—	—	—	—	—	—	—	—
6. SC-2390 × M-197 (TC)	79.00	29.314	10.63	3.13	8.75	5.00	7.03	34.68	28.80	39.88
7. M-197	—	—	—	—	—	—	—	—	—	—
8. Pi 3709 MSC	71.715	23.917	8.13	5.00	10.00	4.36	3.99	31.37	28.42	40.59
9. Pi 3764 MTC	73.286	22.839	8.75	4.38	8.75	4.36	4.53	30.65	27.85	36.44
SD <sub>5%</sub>	0.320	0.134	—	—	—	—	—	—	—	—
SD <sub>1%</sub>	0.430	1.184	—	—	—	—	—	—	—	—

## FORUM

OUR GUEST IS

LÁSZLÓ NÉMETI

Director General

TO THE CENTRE OF STATISTICS AND ECONOMIC ANALYSIS  
OF THE MINISTRY OF AGRICULTURE AND FOOD

PÁL, GY.: *Hungary was historically considered an agricultural country, as the agriculturally cultivable lands amounted to 72 per cent of its total area. This is the best ratio in Europe. The world average is 26 per cent, while in the United States the corresponding proportion is 44 per cent. In this respect Hungary even surpasses Denmark, a country famous for its agriculture. At the same time, Hungarian agriculture shared 12.8 per cent in 1978, 12.3 per cent in 1979 and 11.9 per cent in 1980 from the total investments of the national economy. Don't you think that with such a high proportion of the area suitable for agricultural cultivation (72 per cent) the share of the agriculture (11.9 per cent) from the total investments of national economy is too little?*

NÉMETI, L.: The agriculture plays, indeed, an outstanding role even today in the national economy of Hungary. Large-scale farms and small farmers produce 20 per cent of the national income and employ 19.6 per cent of the labour force, while having a share of mere 12 per cent from the gross fixed assets stock. Thus, as regards its assets supplies level, the agriculture of Hungary is still behind the other branches of national economy and below the international standard. In spite of a considerable progress, the share of the branch from the gross fixed assets stock is smaller than in 1960, 1965 or even in 1975, due to the more rapid development of industry.

Unfortunately the agricultural investments have been realized with considerable fluctuations in time and sum alike.

After the large-scale reorganization of agriculture, the farms shared 16 per cent in 1960, 17 per cent in 1965 and 19 per cent in 1970 from the national economic investments. That was the period of the technical implementation of large-scale farms. Even between 1970 and 1975, large investments — enabling an up-to-date mechanization in the first place — were realized in the branch. It was between 1973 and 1975 that the production systems based on modern technics took shape, making possible the modernization of cereal production and a remarkable increase in its yield level.

From 1975 on, simultaneously with the beginning of a world economic crisis, the development of agriculture has regrettably slowed down, with the result of arresting the extension of large-scale accommodation for animals and machine stock.

The share of the branch from the national economic investments touched bottom in 1980 with 11.9 per cent; then, with the favourable production results of the last two years and the restricted development of other branches, has risen again above 15 per cent.

However, the omission or the restriction of investments in earlier years makes its effects felt in two fields. The fixed assets wear out at a faster rate in agriculture than in other branches of the national economy, so their continual replacement requires rather large sums of money. Today the net value of the machine stock of agriculture is 45 per cent compared to 56 per cent in general industry. The machine stock of the branch, more than 100 thousand million Ft in value, ought to be replaced in this very decade, which with the present sum of 11–12 thousand million Ft allocated a year for machine purchase is impossible.

For the time being, the lack of investments for large-scale accommodation in the livestock branch is a lesser problem, since here the improvement of utilization and specific output rather than an increase in the number of animals is the aim. The development of homeplot farming and small production, and the utilization of former small-scale accommodations for animals also mean a considerable buffer capacity.

The other point of tension is the negligence of plantations. For several years the area of plantations has been decreasing, as the rate of removal exceeds that of replanting.

All in all we can say that investments to back up a more dynamical development of agriculture, a branch of national economy that plays such a great role in the domestic supply and foreign trade balance of Hungary alike, are in any case desirable and necessary.

The investments should, however, be concentrated first of all upon the development of the technical basis, and upon the improvement of the storage capacity serving the preservation of produces, thereby ensuring better marketing conditions.

And with a view to maintaining and widening the small-scale production activity, small machines and technologies required for the modernization of production conditions in this sector must also be provided. In this way, considerable investments from private resources may contribute to the development of the branch.

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PÁL, GY.: *The area of orchards in Hungary was reduced from 156 000 ha in 1978 to 125 000 ha by 1981. The population of the country, on the other hand purchased 7.5 million young trees, propagation material for 8 million currant and raspberry, and 20 million strawberry bushes, and 14 million vine grafts in 5 years. This amount of propagation material corresponds to a plantation of about 20 000 ha. Further, in the homeplots and subsidiary farms there are more than 60 million bearing fruit-trees. In spite of all this the prices of vegetables, and particularly of fruits, are relatively high. Is — in your opinion — this fact due to the labour shortage in the large-scale farms or to the marketing difficulties of homeplots and subsidiary farms?*

NÉMETHI, L.: *In the last ten years the consumption prices of vegetables and fruits rose at the fastest rate, partly for social- and production policies, partly for price policy reasons. The most important of the production policy causes is the fact that live labour is no longer cheap in Hungary. The number of agricultural active earners was halved between 1960 and 1980. At the same time personal income increased to a considerable extent.*

*Since in vegetable and fruit production manual labour is still predominant,*



and its replacement requires great investments, the competitiveness and economic efficiency of these branches are — by the domestic, and especially by the international market standards — questionable or even decidedly low.

It is vegetable and fruit production — and primary production in particular — of all branches of agriculture that have been hardest hit by the price increase in energy, industrial material (pesticides, packing material) and servicing.

The highly labour-intensive and uneconomical branches have become less and less able to compete with the more up-to-date and more profitable branches, and have gradually disappeared from the large-scale farms.

Approaching from the view of price policy, I mention here that the producer's and consumer's prices for agricultural produce and foods of primary importance, from the point of view of life standard policy (some 70 per cent), are officially fixed even today. For a minor proportion of them, the prices are loosely limited, and 15 per cent are free-price goods.

For vegetables and fruits the producer's and consumer's prices are free, so they are determined by current demand and supply, according to the law of the market. The increasing material cost and wages — first of all in the case of direct market goods — can be passed on through the purchase price. On the other hand, this produce does not receive government subsidies, so the consumer's prices really are relatively high.

However, the favourable market conditions and the introduction of the plastic tent method of production made the small producers interested in primary vegetable production. Today the small farmers have a share of more than 56.7 per cent in vegetable, and 80 per cent in primary vegetable production, and their share in grape- and fruit production is also above 50 per cent. In short, the high vegetable and fruit prices are consequences partly of the rapidly increasing material, live labour and servicing costs, partly of the backward production techniques and conditions.

In spite of this, the seasonability of vegetable supply has recently lessened, and these important foods are available — even if at higher prices — almost at any time. The high level of consumption is indicated by the fact that the per capita vegetable consumption in Hungary is today 85 kg, equal to that in Austria and Holland, while the per capita fruit production — due first of all to the rapid growth of apple production — is 30 per cent higher than in 1960, and with its 80 kg quantity is on the same level as those of United States and France.

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PÁL, GY.: Mátyás Matolcsy wrote: "*The results of field crop production by themselves are not sufficient to decide in the question of productivity in favour of the large estate compared to the small holding.*" "*... the intensive livestock farming as well as grape, fruit, etc. production of small farms, namely, fully make up for the loss originating from the lower yield averages of field crops grown in small farms.*" "*... with the annual gross production value per 1 cad. yoke of large estates taken for 100, the value of a year's output from 1 cad-yoke of the small farm is equal to 150.*" What do you think are the present trends of similar indices in homeplots, subsidiary farms and socialist large-scale farms?

NÉMETI, L.: Small farm production plays an important role in agriculture even today. A large proportion of small producers is, however, formed by those homeplots and subsidiary farms which with their production are closely connected with the large-scale farms.

The small producers have the disposal of 12 per cent of the agricultural area, on the whole. (Of this the area of private farms is 1.5 per cent.) However, they occupy

88 per cent of the garden area, 37.9 per cent of the vineyards and 17 per cent of the orchard area of the country. So their importance surpasses by far their territorial proportion, especially in the production of labour intensive crops. While their share in the gross production of agriculture is not more than 33 per cent, in vegetable and fruit production as well as in pig-breeding, they share more than 50 per cent.

The per ha output of small producers — depending on the production structure — is nearly twice as large as that of the large-scale farms. However, the large area output as I have mentioned is due partly to the high proportion — or almost exclusive cultivation — of intensive crops, partly to the fodder provided by the large-scale farm, and in an ever increasing measure to the animals, the breeding material transferred to them by the large-scale farm.

It is perhaps the greatest result (after the large-scale reorganization) of the Hungarian agricultural policy that it has realized the importance of a new form of integration of large and small farms. After 1973 the co-operation between large and small farms became organized.

The large-scale farm invariably has the advantages of carrying out cheap mass production (fodder crops, broiler production, etc.), while the production of a considerable part of the labour intensive crops has been transferred to the small farm.

Small production has even today the advantage of making use of fractions of working time and mostly existing producing capacities. Its activity is characterized by cost and material economy and lower current expenses. The small farm has no obligation of social payments, and its income demand for replacement and development is lower.

We wish to continue to profit by small-scale production in the long run, and we maintain our opinion that the specialized small production — particularly of labour intensive crops — is an important form of commodity production.

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PÁL, GY.: *In comparison to 1970, wine consumption in the 30 largest wine-consuming countries decreased in 10 years by 1.2 per cent. In Hungary the wine consumption has become 8 per cent lower, and is expected to keep on decreasing. The consumption of beer in Hungary increased considerably — by 45 per cent — and of spirits substantially — by 72 per cent — in the same period. The production area of grape-vine was 210 000 ha in 1966–1970 and 206 000 ha in 1970–1975, on the average; then it was 185 000 ha in 1978, 173 800 ha in 1979, 167 800 ha in 1980 and 161 300 in 1981.*

*Is, in your opinion, the reduction of the grape-vine area the cause for the decrease in wine consumption, or is there no connection between the two phenomena?*

NÉMETHI, L.: Changes in the consumption habits parallel with a rise of the life standards must be considered natural. So, some decrease in the wine consumption cannot be regarded as unfavourable. The change in the consumption structure in favour of beer consumption — that has increased to 88 litre per capita, more than tenfold compared to 1950 — is thought to be favourable. As to per capita beer consumption, we have caught up with Holland, which means that we no longer fall behind.

The per capita wine consumption in Hungary rose from 33 litres in 1950 to 37.7 litres by 1970, then fell back to 34 litres by 1980. Nevertheless, Hungary exceeds most European countries in wine consumption — except the large wine producers — and is on the same level with Austria.

In the decrease of wine consumption, some role was played — in my opinion —



by the fact that the primary cost and consumer's price in the highly investment and manual labour intensive branch of wine production increased at a faster rate than the cost and price of beer and spirits, that are cheaper for big industry to produce. The consumer's price ratio has thus changed in favour of beer. This process was promoted by the efforts made in the last decade to improve the quality of wine, to produce quality wines. These efforts are partly justified by export interests, and by better marketing possibilities of quality wines.

Even our variety policy tended to satisfy this demand. So the cheaper mass wines have disappeared from production with the result that, in spite of the increasing yields and saturated market supply, the domestic wine consumption of Hungary decreased because of the high prices.

The wine production of Hungary — with the record yield of the last year — rose far above the earlier level. The quality of wine is also satisfactory. The decrease of consumption can thus be explained exclusively with a change in the consumption habits and the relatively high consumer's prices.

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PÁL, GY.: *According to the general opinion, the level of university education in technical sciences is higher in Hungary than required for those employed in industrial units. In the field of agricultural sciences, on the other hand, university education is on a lower level than demanded by the agricultural enterprises. However, among the managers of industrial units the number of graduates is much smaller than in agricultural establishments, where their proportion is considerable. What do you think, is it a smaller number of highly educated or a larger number of less qualified leaders that can successfully increase production and economic efficiency?*

NÉMETI, L.: The question is very interesting and touches upon a timely problem. I am convinced that under the present developed, or even elaborate conditions of production, the role of professional training is becoming more and more important. I emphasize that it is not only the leaders or university people but the entire collective taking part in production, that I think of here.

In the last fifteen years it was undoubtedly in agriculture that the economy of Hungary made the greatest progress. Agriculture has succeeded in synthesizing the most up-to-date varieties and production technology of the world, and using them efficiently in an organic system. This complex development and progress — a switch-over to new technics — has taken place without any considerable losses.

No doubt, it was a matter of adaptation in the first place, and — I think — that is why it was successful. However, the fact that the workers after a short training were able to cope with the most complicated technics proves better than anything their aptitude, versatility and enthusiasm. Of course, material incentives and the possibility of showing off were also necessary. In agriculture it was natural that the best trained and cleverest workers were put on the new machines, and in proportion with their performance received higher wages and recognition. At the beginning a good combine operator often was given four times the average wage during the harvest, sometimes more than the first leader of the unit.

Thus, I think, the source of production successes is a good staff of skilled workers.

The school training of production organizers and managers was occasionally — first of all at the time of the rapid technical instrumentation of agriculture — not as



good as desired, but young people put to work in a farm mostly are immediately given independent scopes of activity, and thus have the opportunity of quick acclimatization.

Production organization usually depends today on the precise implementation of an established technological order. The task is mostly accomplished by the professional staff on an appropriate level.

Production development, variety supply and machine testing generally are not the tasks of the operative staff, — moreover, mostly not even of Hungarian experts. The professional staff of the agricultural establishments satisfies the demand of production organization, as proved by the remarkable production successes.

The deficiency of training is felt in the sphere of economic management and guidance rather than in the field of production.

It must be noted that the new technics, the change of variety, the independent enterprise management demand highly qualified experts first of all on the level of leadership. In this respect, university education of recent years and the activity of the graduates do not fully meet the requirements. Consequently, the up-to-date instruments of production are not joined by a technical basis and operative demand that would enable management the analysis of management (the consumption of nutrients, fodder, fuel and machine components) with the help of which a higher level of economical management could be attained.

The same applies to the enterprise level management, where appropriate analyses and marketing activity are still not carried on. Naturally, this requires the improvement of guidance as well. Here it should be mentioned that there are even today great differences in the level of management work. An ever-increasing number of farms in Hungary are managed on a high level by well trained leaders who employ up-to-date methods, while in many farms — first of all in those with unfavourable natural — or economic conditions — the professional staff is objectionable both from quantitative and qualitative points of view.

It is thus in these fields that the level of professional education should be first raised. I am convinced, however, that in possession of adequate conditions and instruments (for example, computers) enterprise management can also be rapidly improved.

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PÁL, GY.: *Precipitation in the vegetation period (from 1 April to 30 September) ranges between 300 and 350 mm in the larger half of the area of Hungary, and in 3 of 4 years only 275–300 mm precipitation can be reckoned with in the central part of the Great Hungarian Plain. At the end of the 1940s, 70 per cent of the irrigation area covered low fertility soils poor in precipitation, mainly because of rice production. Irrigation — on a farm level — was more efficient on higher fertility soils, since the economic efficiency indices of irrigation rise and decline with the fertility of soils. Is it — in your opinion — on poor or on better quality soils that irrigation should be first developed?*

NÉMETI, L.: I think the question can today be unambiguously answered. The experiences and analyses of the last years prove that — considering the high expenses of irrigation — the better quality soils are primarily worth being irrigated. As opposed to the conception of an earlier period — when the poor quality fields were utilized by rice production — this applies to rice production as well. Even with rice, high yield can be obtained only on good soils. For some years the irrigation of grasslands too has considerable increased; in 1982 grasslands occupied some one-third of the irrigation area. However, the extension of irrigation, here too, took place on areas with better quality soils.

This tendency is indicated by the fact that irrigation systems are being constructed first of all on good quality lands, and in farms with high management levels.

It should be noted, however, that the shift of the production structure of agriculture towards the cereal crops, the reduction of the proportion of large-scale vegetable production and other intensive crops, act against the extension of irrigation on these areas too. It must also be taken into consideration that, with the present up-to-date varieties and technology, high yield averages can be attained by chemical methods even in dry farming.

The high costs of irrigation, the expensive main works and irrigation systems, and the raised water-charges, have resulted in a considerable regress of irrigation; today the costs of irrigation are seldom returned by a surplus yield. Therefore high irrigation capacities — the main works in the first place — are now left unexploited.

Of the agricultural area of Hungary, only 2.4 per cent is irrigated today, and this proportion is not even expected to grow in any considerable measure. However, simultaneously with the reduction of irrigation the demand for a complex melioration has increased, with the view of improving the fertility and production reliability of areas with unfavourable water conditions.

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PÁL, GY.: *In 1981 the export choice of the Hungarian national economy consisted of 1670 kinds of goods. More than half of them — 850 — had a mere 2 per cent share from the total value of exports. These data show that the foreign trade returns come decisively from traditional products well established on the markets and sold in bulk. However, it is these very goods of which selling on capitalist markets closed by various discriminations is made difficult by the economic depression.*

*What do you think; is it in the large-scale farms or in the homeplots and subsidiary farms that the export choice of the export oriented Hungarian food economy can be better widened?*

NÉMETI, L.: I think the future of Hungarian food exports is more complicated than that. For some years, the world market possibilities for food have not improved, in spite of the increased volume, national economic share and balance-improving role of our food exports. The increasing market competition and the prices are determined — more definitely than ever — by countries capable of cheap mass production (for cereals: the United States, Canada and Argentina; for meat: Austria, New-Zealand and Argentina). Our traditional markets for vegetable and fruit, on the other hand, are threatened by the South-European countries of more favourable climate. Besides, the soundly developed European markets are characterized by high protective tariffs and a tendency to self-sufficiency, while on the socialist markets only cheap bulk goods can be sold even today. The markets of the Near-East are characterized by a great uncertainty and a keen competition.

Our export structure must thus be adjusted to the demands and conditions. Accordingly, the exports of grain crops, vegetable oil and pork are invariably favourable; while vegetables and fruits, poultry and mutton are more and more difficult to sell, either in fresh or in processed form.

The products of the cereals and meat branches have today a 65 per cent, the horticultural produces a 26.5 per cent share in the export returns of Hungary. Sixty per cent of the goods was sold in a processed form, and 72 per cent of them for dollars.



Production and export policies should therefore be primarily adjusted to the demands. And the demands are highly differentiated. It is naturally in the large-scale farms that the economical conditions of cereal production — which also forms the basis of livestock farming — are present. The same applies to vegetable oil and seed production.

The small farms have a considerable share in the export commodity production of pork, poultry, and — mainly for reasons of quality requirements — of labour intensive fruits, first of all berries.

Further, the small-scale farm plays an important role in the production of other small animals (80 per cent) which are also significant as sources of returns.

In producing the commodity funds of exports, an important part is thus assigned to the small producers.

However, according to the law of the market, uniform bulk goods are necessary to keep a market in the long run. Permanent and reliable sales require the stabilization of marketing conditions, a possibly direct wholesale marketing of great bulks of uniform goods, and almost daily deliveries. This naturally does not exclude, but renders it decidedly necessary, to make use of the qualitative advantages of small-scale methods in production, and occasionally in packing. The integration solutions of the large-scale farms may be the main forms of this.

So, in my opinion every sector, moreover the whole vertical system of production and marketing, has its function in a better utilization of our export possibilities.

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PÁL, GY.: *According to the UNO surveys not only the demographic changes caused by the war but also the economic development and the change of the manner of life are responsible in Europe for the birth rate having fallen below the reproduction level in Austria, Belgium, Denmark, Finland, France, Holland, Great Britain, German Federal Republic, Norway, Switzerland and Sweden. The experts unanimously are of the opinion that it is poverty and not welfare that brings the large family into existence. According to the demographic forecasts the number of population in Hungary will be reduced by one and a quarter of million by 2021. How many people could be fed by the Hungarian food economy?*

NÉMETHI, L.: The Hungarian food economy is even today capable of remarkable achievements. Besides an appropriate level provisioning for the ten and a half million inhabitants of the country, some one-third of our food production is sold abroad. That is, food production in Hungary is at present capable of supplying 15–16 million persons with food on a high level. And as for the most important foodstuffs, the ratio is much more favourable than that. For example, our domestic production of grains, meat and eggs, as well as of vegetables and fruit renders it possible to purvey provisions for about 20 million people even today. Considering the number of those engaged in food production, on the other hand, each one produces food for 18 persons.

We can reckon with a further considerable increase in the production and productivity of the branch even if we take into account the changes predicted until the end of the century — the decreasing production area and the slowing rate of development. Thus, the grain crop production of Hungary may rise by some 25 per cent to 18 million tons, the meat production by 20 per cent to 2 million 400 thousand tons. Egg production may also grow by about 50 per cent. The vegetable, fruit and grape production, on the other hand, will not increase much.

In accordance with the life standard policy and export demands, food processing



will be increased at a faster rate than the growth of agricultural production, and the preparation of products will considerably improve.

With a total of some 50 per cent growth of food production until the turn of the millenary, and the rising level of domestic consumption also taken into consideration — which may mean an increase of about 10–15 per cent — the sustaining capacity of the Hungarian food production is estimated to be about 20 million persons. In the meantime the number of workers engaged in agricultural production and food processing will decrease by a further 30 per cent or so; at the turn of the millenary, one worker employed in food production will produce an amount of food required for 33 persons.

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PÁL, GY.: *Among the capitalist countries it is only in Argentina that the producer's price for wheat is lower than in Hungary; in the United States, a country with a decisive influence on world production, the price level is mostly similar to Hungary's, and sometimes 10–16 per cent higher. In Denmark and France it is twice as high; in Austria, Holland and the German Federal Republic and Italy two and a half times as high as the Hungarian wheat price. In the German Democratic Republic it is 20 per cent lower; in Poland, 56 per cent higher. In Holland 0.37 tons, in Hungary 0.95 tons of wheat must be given for 1 ton of fuel oil. Will the Hungarian agriculture be capable of reproduction on an increased scale with the present price ratios and the regulation system in effect?*

NÉMETHI, L.: The question of price ratios has always been particularly important in Hungarian agriculture. It is a well-known fact that the Hungarian price ratios have for decades been unfavourable for agriculture. Even after the economic reform introduced in 1968, a considerable price disparity prevailed, which, however, was mostly compensated for by various subsidies until 1975, so that in essentially the agricultural price gap did not change in that period.

In the years following 1975, on the other hand, with a more rapid rise in the industrial prices the agricultural price gap became wider. After 1980 this process accelerated, so that in 1981 the agricultural producer's prices were higher by 53 per cent, and the prices of industrial goods used by the agriculture by 77 per cent than in 1970. In 1982, then in 1983, further curtailments came into force: the reduction of subsidies, and the increased obligations of paying in, worsened the income position of the branch.

The growing burdens of interest and the reduced time of repayment also decrease development funds and the possibility of reproduction on an increased scale, respectively.

It is a further problem that in spite of the record yields of the last year the profitability of the agricultural production and — owing to the regulatory measures — the possibility of reproduction on a greater scale decreased. According to our investigations the differentiation of farms continued in 1982. As a result, 40 per cent of the state farms dispose of 10.6 per cent, while 25 per cent of the co-operative farms of 5.5 per cent of the development funds of the sector. Taking also into consideration that these are the very farms where the level of instrumentation is the lowest, the stock of assets badly worn away, and the development funds severely charged with credit repayments, we can draw the conclusion that even a simple replacement of their assets is difficult to carry out. Some 40 per cent of the large-scale farms are incapable of reproduction on an increased scale, so a further reduction of their net stock of assets can be expected. Even within the remaining 60 per cent, only about

15–20 per cent dispose of any considerable resources that render the development possible.

A further danger may be represented by the fact that, owing to the low profitability of agricultural activity, the industrial and servicing activities which temporarily ensure higher incomes are given priority when the investment resources are allocated. In this way a further regrouping of the agricultural capital may take place with the possible result of additional restrictions in the agricultural production of large-scale farms (first of all in the investment of intensive cattle farming and plantations).

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PÁL, GY.: *It was in 1982 that major difficulties of selling first arose in Hungarian agriculture. Lamb, sheep and poultry were the first to cause problems, followed by fruit; but selling grapes and wine was also difficult. Owing to the problem of selling the amount of commodity produced it is to be feared that production of these goods will decline; because, for example, the farms producing poultry could not deliver their live animals at the time fixed by contract, had to keep them longer and the cost of keeping was charged to them. Are — in your opinion — these phenomena the first signs of an agricultural overproduction?*

NÉMETHI, L.: Although the facts mentioned in the question are undisputable, I do not think that an agricultural overproduction could be spoken of. It is in any case a paradoxical situation that we have marketing problems when there are considerable shortages of food in neighbouring countries.

In my opinion it is a decrease in sound demand more than anything that overproduction is related with, which equally occurs in the developed capitalist countries and in the socialist countries. Beyond a reduction of food consumption and demand, declining or stagnating living standards involve a decrease in the demand for quality and increase in that for cheap mass products.

The conception propagated in the past period: to produce everywhere saleable goods has turned to its wrong side. The quality of our goods — due first of all to the way of processing and packing — does not reach the level required by the developed countries and is therefore less competitive there; while, on the socialist market, quality is not much recognized by the price.

As I have already mentioned, competition on the food market regrettably increases. Developed capitalist countries, and developing countries too — the latter with considerable subsidies — sell their agricultural produces at extremely low, dumping prices. This applies today to cereals, vegetables, fruit, wine, poultry, eggs. At the same time the developed capitalist countries — to protect their producers — impose high protective tariffs on imported goods. In this keen competition we, too, are only able to keep our markets with a considerable state subsidization.

For some years, however, owing to the faster increase of industrial prices and costs, the cost of domestic production has grown with a simultaneous increase in the risk of sales. The position of the agricultural producers is made difficult by the fact that the risk of production and selling is largely shifted upon them.

One of the unsolved problems of our economic management is that the producing farm is separated from the market, and it is usually the foreign trade enterprise or the budget that profit by the sales, while the losses are mostly suffered by the producer.

With a view to the reliability of production, the risks must be taken in common, and an active operation of reserve and equalizing funds is demanded.

It has to be taken into consideration here that for large-scale production — on account of specialization — the reliability of marketing is much more important. A failure of sales or readjustment of production would occasionally result in considerable operative or even social losses. That is why organized and lasting market relations and the right production structure shaped for a long period ahead are so important.

From the point of view of the market flexibility of our economy, important reserves are — in my opinion — represented by the small-scale production, which for the very reason of its lower assets requirements and higher mobility of live labour and management is capable of quick adaptation.

For small producers specialized in commodity production, the reliability of production is also of increasing importance, since a modification of the structure is — here too — more and more expensive.

I think thus, that the safety of production and producers can be ensured by reducing the costs of production, increasing the market relations and interests, and making the system of various equalizing and reserve funds or budget subsidies more flexible.

However, a selective development of the production structure, better adjusted to the demands, is also necessary. Branches and vertical systems representing the bulk of our export goods should be marked out and their interest systems stabilized for a longer period, while with products of lower importance a greater enterprise, market and interest mobility should be allowed. So, the production of cereals and meat — first of all pork — should be encouraged, while vegetable, fruit and poultry production, in which the competitiveness of Hungary is difficult to improve, restricted.

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PÁL, GY.: *Thank you for the information.*





## RECENSIONES

*Monoufeia Journal of Agricultural Research*. Vol. 1-3, Published by Faculty of Agriculture, Monoufeia University, Shebin el-Kom, Egypt, 1978-1980

Shebin el-Kom is the centre of Monoufeia Province, one of the most important agricultural areas in the delta region of the Nile. The Faculty of Agriculture at its famous university launched a new English yearbook-type journal, using stencil presswork.

The editorial board in 1980 consisted of Prof. Dr. Mohamed El-Kady, Prof. Dr. Mohamed N. Shatla (editor) and Dr. Mahir A. Nawar in 1978, and of Prof. Dr. Mohamed El-Kady, Prof. Dr. Nahir A. Nawar (editor) and Prof. Dr. Aly F. Omran.

The first volume runs to 402, the second to 405 and the third to 445 pages. Structurally the articles follow the usual international pattern except that the literary references precede the tables and figures; and finally, each article is completed by a brief, one-page summary in Arabic. The three volumes deal with 68 subjects in the form of articles occasionally consisting of several parts. A publication of economic subjects was written by the authors in Arabic.

On the basis of their titles and contents, the articles listed below can be placed in the following subject groups:

### 1. Soil Science

Effect of certain granular insecticides on the nitrifying power and *Azotobacter* in cotton-cropped soil

Effect of organic matter and seasonal conditions on the behaviour of some soil pesticides in soil and plant

Effect of soil moisture and seasonal conditions on the behaviour of some soil pesticides in soil and plant

Land relief and its effect on soil salinization and alkalinization of the Fayoum depression

Reclamation of sodic-saline soil by amendments and electricity

Some factors affecting adsorption of  $\text{Na}^+$  and  $\text{Ca}^{++}$  by excised roots of maize

The effect of saline irrigation water of different SAR on some properties of certain alkali soils

The influence of saline water on denitrification in soil

The side effect of some soil pesticides on kidney bean plants under various conditions of organic matter, soil moisture and seasonal effect

### 2. Effect of nutritive elements on cultivated plants

A comparative study on the effectiveness of NPK soil fertilization and macro- and microelement foliar fertilization on yield and quality of Romi-Red *grape* variety

Effect of plant density and nitrogenous fertilizer on *okra* plants. I. Growth and chemical contents. II. Yield and pod characteristics

Effect of phosphate, as localized on foliar applications, and nitrogenous fertilizer along with their interactions on snap *bean* plants. I. Growth and chemical constituents. II. Intake rate and efficiency of utilization of phosphate and nitrogen. III. Yield and yield components

Effect of some fertilizer treatments and previous crop on growth and yield in *rice*

Growth and essential oil production of *Ocimum basilicum* in relation to nitrogen and phosphorus fertilizers. I. Growth and chemical constituents. II. Essential oil production

Yield and fruit quality of Biz-Elanza *grapevines* as affected by foliar fertilization

Yield response of *onion* to some fertilizers

### 3. Application of growth regulators in crop production

Comparative studies on yield of *tomato* cultivars treated with various growth regulators

Effect of gibberellic acid, alar and ethep on some chemical characteristics of *plum* fruit

Effect of gibberellic acid on the growth and flowering of Queen Elizabeth and Bacara rose varieties

Effect of 2-chloroethyl trimethyl ammonium chloride (Cycocel) on yield and grape quality

Effect of some growth regulators on growth, flowering and yield of *Datura* plant

Effect of some growth regulators on internode length of new shoots, flowering and fruit abscission of plums

Effect of various growth regulators on some enzyme activities and phenolic compounds in *Datura* plant

Preliminary studies on the effect of gibberellic acid on the growth and flowering of carnation plant, *Dianthus caryophyllus* L.

Response of wheat plants to growth regulating substances in combination with some macronutrients

The effect of some growth regulators on yield and fruit quality of Romi-Red grapevine

#### 4. Agrotechnics, mechanics

Effect of some agricultural practices on the growth and yield of some Egyptian cotton cultivars

Effect of transplanting date, plane density and cultivars on yield and quality of onion (*Allium cepa* L.)

Plant population in relation to growth, fruits dimension and yield of cucumber

The effects of weed control by Cazaron on shoot production of Italian grapevines

Theoretical studies on some open belt drive systems

#### 5. Plant breeding, -genetics

Combining ability studies in a five parent diallele cross in common wheat including relative contribution of flag leaf

Effects of ethyl methanesulfonate and N-methyl-N-nitro-N-nitrosoguanidine on *Allium cepa*

Heterosis in a five parent diallele cross in common wheat including relative contribution of flag leaf

#### 6. Phytopathology, plant protection resistance

Effect of several fungicides on yield and quality of onion

Effect of soil tape on *Meloidogyne javanica* population in cowpea roots in relation to plant growth and nutrient uptake

Efficiency of certain nematocides against the stunt nematode (*Tylenchorhynchus latus*) on cotton

Efficiency of some insecticides in controlling the bean pod-borer *Etiella zinckenella* Treit. on leguminous crops

Interaction between root-knot nematode, *Meloidogyne javanica* and soil acari in relation to horse-bean nematode infection

Interrelationships between plant-parasitic nematodes and soil acari. I. Interaction between root-knot nematode, *Meloidogyne javanica* and soil acari in relation to tomato infection

Population density of the tarsonemine mites under certain truck crops (Acari: Tarsonemina Pygmephoroidae, Tarsonemoidae)

Preliminary observations on the population density of mites inhabiting vegetative parts and soil of certain wild plants at new reclaimed lands

Races of stem rust, *Puccinia graminis* f. sp. *tritici* in Egypt during 1974-1976

Studies on *Drechslera australiensis* and *Alternaria alternata* leaf spot of soybean in Egypt

Studies on the nature of disease resistance against purple blotch infection of onion

Varietal susceptibility of tomatoes to root-knot infection

Vertical distribution of the tarsonemine mites under wheat in Monoufeia governorate

#### 7. Microbiology, enzyme production

Chemical and bacteriological studies of some lactic cultures in fermented milks

Factors affecting microbial cellulase production from cereal by-products. I. Effect of environmental conditions. II. Effect of nutritional conditions

Studies on aflatoxin production by different *Aspergilli* isolates from soybean

Studies on proteolytic and lipolytic microorganisms isolated from deteriorated old leather manuscripts

Studies on *Sclerotium cepivorum* Berk toxins

Survey of microorganisms for the production of cellulase enzymes from cereal by-products

The inhibitory effect of some fermented milk flora on the growth of some pathogenic bacteria

#### 8. Animal breeding and processing feeding

A study of body weight and measurements on commercial Ossimi flocks of sheep raised at Monoufeia province

Chemical and organoleptic evaluation of hamburger and canned-type product containing lean meat mixed with soybean and chick-pea

Effect of feeding lactating ewes urea ration on some microbiological properties of Dani cheese

Effect of pepsin treatment on some chem-



ical indices of bastirma processed from *camel* meat

Free amino acids contents of *camel* meat as influenced by pepsin, bastirma processing and storage

Low temperature preservation of *rabbit* embryos in plastic straws

Ovarian development in Egyptian *buffalo* heifers

Physical properties and protein solubility of bastirma prepared from *camel* meat tenderized with pepsin

Reproductive performance of the commercial Ossimi flocks of *sheep* in Monoufeia province in Egypt

The effect of replacing fish meal with a part of soybeans or decorticated cottonseed meal on fertility, hatchability and egg production of *foyoumi hens*

The influence of feeding different concentrate to roughage ratios on growth of male and female *cow* calves

#### 9. Economics

Changing cost and profitability for selected commodities in Monoufeia Governorate and Egypt

Forecasting model of grain production in relation to weather variability in Morocco

Marketing margins for selected commodities in Monoufeia Governorate and Egypt

As regards their contents the publications reach the world standard. The research results they contain not only concern Egypt; there are novelties in them that may attract international interest. The Hungarian and foreign agricultural libraries would be incomplete without the Monoufeia Journal of Agricultural Research.

L. GY. SZABÓ

*Physiological Plant Ecology I, Responses to the Physical Environment*. Edited by O. L. LANGE, P. S. NOBEL, C. B. OSMOND and H. ZIEGLER (Encyclopedia of Plant Physiology, New Series, Vol. 12A). Springer-Verlag, Berlin—Heidelberg—New York, 1981. 110 figures, 625 pages.

By publication of this work, Springer-Verlag has completed its deservedly popular *Encyclopedia of Plant Physiology* with a new range of subjects. This issue summarizing and analysing physiological plant ecology, has a pioneer aspect in the literature, since the definition of this branch of science itself dates back only a few decades. Although its elements existed within the interdisciplinary nature of both plant physiology and plant ecology, its development into an independent branch of science could only begin in the

early 1970's. What, after all, is meant by physiological plant ecology? According to the brief definition in a Hungarian university textbook, "... analyses of physiological processes with ecological objectives are specified as eco-physiology". The definition given in the introduction of this volume, though much more concrete, will certainly need further completion; "The objective of ecological plant physiology is to explain processes in plant ecology, such as plant performance, survival, and distribution, in physiological, biophysical, biochemical, and molecular terms". The affinity of the Springer-Verlag Publishers to the research development is shown by the fact that they also published the first English summary on the subject (Larcher 1975). The success of their publication seven years ago will certainly be surpassed by this more substantial and up-to-date book, guaranteed by the participation of W. Larcher and the team of well-known authors who compiled the volume.

This is the first of a projected four-volume series:

- I. Responses to the physical environment
- II. Water relations and carbon assimilation
- III. Responses to the chemical and biological environment
- IV. Ecosystem processes: Mineral cycling, productivity and man's influence

The reader is greatly helped by the information included on the current subjects and even on the chapters of the future three volumes.

The range of subjects indicates the complexity of the task undertaken by the publishers. In such a case it is inevitable to compromise for the magnitude of the work by separating the first from the second volume, although the water supply and its effects could probably have been included among the system of physical factors. Similarly, the separation of some chapters of the third and fourth volumes may neither have been a simple task for the editors.

This first volume of the series discusses in 17 chapters the role and influence of physical factors. In chapter one (Fundamentals of radiation and temperature relations) an up-to-date survey of the radiation relations essential for the life processes of plants is given, as well as the energy turnover of the plants and its effects within the canopy. However, the energy balance and utilization, of a single model plant are easy to interpret. In the case of plant canopy, many "disturbing" factors occur; and processes taking place inside the tissues are equally difficult to follow. In agreement with the author of the

chapter, the extension of knowledge in this field is definitely required.

Chapter two describes the photosynthetically active radiation. Its importance has increased in interpretation of processes taking place in nature as well as in laboratory analyses (under controlled conditions) and glasshouse production. However, the chapter indicates that neither the measuring of PAR nor its definition are easy tasks. This is confirmed by the excellent material compiled on the radiometric photon flux measuring of PAR, on the physiological reaction and on errors of measuring.

An excellent summary of the eco-physiological effect of different quantum flux densities is given in chapter three (Responses to different quantum flux densities). The influence of light supply is presented by the synthesis of results attained in the last decade. Thus the chapter analyses the physiological differences of  $C_3$  and  $C_4$  species, the changes of enzyme reactions, the questions of electron transport and photoinhibition. The value of the chapter is enhanced by the numerous measuring data and by some speculative theory concerning the part processes.

Increasing attention has lately been paid to the effects of spectral composition, the spectral energy distribution of light on the non-photosynthetic processes. Its importance has been realized, moreover, the indirect effect of spectral composition on photosynthesis is not negligible either. These points are discussed in chapter four (Non-photosynthetic responses to light quality) and chapter five (Responses to photoperiod). The separation of this subject in two chapters seems somewhat arbitrary, since chapter four devotes equal mention to all main directions of action, such as photomorphogenesis, phototropism and photoperiod. The separate discussion of photoperiodicity can only be justified by the wider range of information available. However, both chapters supply a high level of the most current information.

Chapters six and seven (Plant response to solar ultraviolet radiation, Responses to ionizing radiation) analyse the effects upon the plant of the two special wave lengths of solar and artificial radiation. In discussing the effect of UV radiation, this chapter acquaints the reader with the sources of radiation and its spectral activity, with the diversity of plant responses and patterns of plant adaptation. Such emphatic discussion of ionizing radiation cannot be regarded as superfluous. A much new information is included concerning tolerance, sensitivity, genetic responsiveness, etc., of plants. The description of the adverse effects of "man-made" ionizing radiation, caused by nuclear weaponry, can be regarded as a warning.

In chapters eight and nine (The aquatic environment, Responses to light in aquatic plant) the reader is introduced to a special field of eco-physiology. Chapter nine concentrates upon discussing water movement, nutrient turnover, and dry matter transport of aquatic plants. Starting from the description of special features of aquatic plants, the question of photic environment, the characteristics of the special pigment system and the physiologically active radiation forms are also examined.

The subjects of the next five chapters pertain to the problem of temperature. Temperature has extreme eco-physiological importance in the distribution of plants, and modification of their production processes. Thus, the description of the plant responses to temperature constitutes the longest part of the book (Responses of Macrophytes to temperature, Responses of microorganisms to temperature). The question may arise whether the separation of the subject into macro- and microorganisms (chapters ten and eleven) was really necessary. Otherwise, such interesting as the temperature of tissues and cells, physiological and biochemical responses to the effect of temperature, the distribution of plants, etc., are discussed in detail. The description of thermoregulation, and the analysis of the adaption of microorganisms and their tolerance to extreme temperatures are also valuable parts of this chapter.

Chapter twelve (Responses to extreme temperatures. Cellular and sub-cellular bases) is devoted to three main topics; high temperature injury, chilling injury and freezing injury. The complex nature of these is shown by the fact that the author separates the material in to 28 sub-chapters. Unfortunately, some sub-chapters offer only brief information, obviously because of the restrictions of space.

In chapters thirteen and fourteen the ecological role of low and extremely high temperatures is discussed, using the clearest eco-physiological attitude (Ecological significance of resistance to high temperature). The well-constructed tables and figures are of great help in familiarizing readers with the subject. The most recent discoveries concerning plant adaption, stress effects, distribution of plants and control of physiological processes are presented on a high professional level.

In the last few years, investigations into the ecological role of wind have become more frequent. Consequently the subject is separately discussed in chapter fifteen (Wind as an ecological factor). However, the topic of air composition receives less attention than a more detailed analysis of the effect of air motion. Here, the reader is acquainted with



the plant atmosphere and surface phenomena, the direct mechanical and physical actions of wind and its indirect biological role, such as transportation of pollen, spores, reproductive organs, etc. The review of the influence of wind on growth and development is also extremely interesting.

Fire as an ecological factor is restricted to only certain areas and vegetation types. For this reason, it is rarely or very briefly discussed by European authors. In spite of this, chapter sixteen (Fire as an ecological factor) contain many remarkable details which may arouse every plant biologist's interest. To mention but a single example, the stimulatory effect of fire on germination, growth and development is discussed.

The last chapter of the book (The soil environment) is one of the shortest in the volume. It is highly regrettably because the soil is the most complex and at the same time the most synthetic ecological system of all. Consequently, the information about it must be rather deficient. However, according to the list of contents in the future volumes, a more detailed discussion of the subject can be expected. It is one more reason to hope for their publication as soon as possible.

Despite some of these remarks, the book is the best existing publication on its subject. It must be emphasized that not only the synthetic analysis of information is new, but it can also be considered the first complex formulation of plant eco-physiology that will determine research directions in future years. I am convinced that the book will be regularly used in the everyday work of plant biologists.

J. BERNÁTH

*Physiological Plant Ecology II. Water Relations and Carbon Assimilation.* Edited by O. L. Lange, P. S. Nobel, C. B. Osmond and H. Ziegler (Encyclopedia of Plant Physiology, New Series, Vol. 12B). Springer-Verlag, Berlin—Heidelberg—New York, 1982. 153 figures, 747 pages

The Publishing House of Springer-Verlag, quickly fulfilling its promise, published the second volume of its ecophysiological series within a year. This surprisingly short time indicates the ever accelerating process of obtaining and publishing new scientific results. Let us remember this volume of the Encyclopedia of Plant Physiology on a similar subject, the description of water regime in plants (Water Relations of Plants, 1956) was followed by the volume containing the treatment of carbon dioxide assimilation (The Assimilation of Carbon Dioxide) only four years later. It may, naturally, occur to the

reader, irrespective of the thoughts in the introduction, to ask the reason for combining water relations and carbon assimilation — two seemingly very disconnected subjects. The arguments listed in the introduction are convincing, even if from a different point of view reasons may be given for their separate discussion. Nevertheless, it can hardly be denied that for some ten years the researchers have paid special attention to the water status and carbon dioxide turnover of plants, and to the ecotypic differences shown in them. These facts were established on the basis of recent investigations; the relationship between water regime and photosynthesis in the  $C_3$  and  $C_4$  species, and in the CAM plants are in any case strong arguments for the publication of the material in this form.

The book is divided into 18 chapters and is made complete by an index of authors, and a plant name and subject index. As a new aspect in Chapter 1 (*Water in the Soil-Plant-Atmosphere Continuum*) the water regime and production of the plant is viewed over a long term period. Namely, by water regime a shorter interval or even a given point of time (status) was indicated earlier, while the production was referred to a vegetation period. Now the water appears in different phases of vegetation as a limiting factor, in interaction with the adaptation of the plant. Chapter 2 (*Water in Tissues and Cells*), on the other hand, discusses the water status of the plant and its physiological questions in a given point of time on the level of tissues and cells. Such often discussed questions as the "matric" and "osmotic" potential come into prominence, but the properties of the cell-wall responding elastically or plastically to changes in the water status are examined anew. The chapter even offers a method for measuring the latter changes. The next part 3 (*Water Uptake and Flow in Roots*) further develops the logical progress of the book. Although there are some familiar questions here, the formulation of the material is new, and the hypotheses are supported — and thus made acceptable — by numerous data. The material is centered around three basic questions: a) the hydraulic resistance of the root, b) the pathway through the root, c) the hydraulic resistance of the soil up to the root surface. Water uptake through the root is not, however, the only pathway in the plant world. It is a relatively familiar fact that other plant organs are also able to assimilate water. In this respect the differences among the species, the form of ecological adaptation are, naturally, of decisive importance. Attention is called to by the large number of new data in Chapter 4 (*Water Uptake by Organs Other Than Roots*).



Of course, a better understanding of the mechanism of this form of water uptake, and of its role in the ecological adaptation requires — as pointed out by the author — the wide application of up-to-date means of the ecophysiology of plant. Chapter 5 (*Transport and Storage of Water*) describes what happens to the water in the plant. The continuity of water transport through the conducting units, tracheids and vessels is ensured. The system and mechanism of water transport are adapted to the ecological conditions, so it is by all means justified to discuss them from an ecophysiological point of view. The storage of water is also of ecological importance. The accumulation of water in the xylem, in the bark or in the primary tissues, requires different forms of adaptation. In Chapters 6–8 the same scope of subjects is treated, and the ecophysiological regularities of water discharge. The first chapter on this group of subjects, compiled admirably, discusses the process of water discharge within the tissue and between the cells. From an ecological point of view, the water retention of the cuticle is important. It depends — according to the facts listed by the author — on its structure rather than, as generally thought, on its thickness. It remains, naturally, an open question what the connection between water permeability and the amount of soluble lipids present in the cuticle is. Transpiration through the stomata and  $\text{CO}_2$  uptake related with it are summarized on an outstandingly high level, based upon results attained in the last decade, in Chapter 7. Chapter 8 provides the mathematical model for this group of questions.

A new, though related scope of questions is covered by the next three chapters of the book: *Physiological Responses to Moderate Water Stress, Desiccation-Tolerance, Frost-Drought and Its Ecological Significance*. The effects of a deficient or critically extreme water status and the forms of responses by the plants are set forth on the basis of the most recent findings, and no secret is made of the fact that many details of the mechanism still remain in question. New light is thrown upon the connection between stress and synthesis of growth regulators, relations of hormonal control, and changes in the RNA-protein metabolism in response to desiccation.

Chapters 12–13 discuss the ecophysiological relation of an exactly defined life process: seed and spore germination to water. The results achieved in this field during the last two decades are summed up on the basis of nearly fifty literary citations. Beside analysing the physiological role of water the chapter also touches upon the process of water uptake (question of membrane, energetical implications, etc.) and upon the ecophysio-

logical control exercised by the water on germination. However, the character of this subject deviates from the conception indicated in the title; exploration of the connection between water and carbon dioxide assimilation. It is all the more so with Chapter 14 (*Physiological Responses to Flooding*). This is only an uncritical remark, since the chapter treats a little known scope of subject on an up-to-date level, as indicated by the fact that most of the nearly two hundred literary sources cited were published in the last ten years.

It is in the last four chapters that the editors' basic conception is crystallized. In Chapter 15 (*Functional Significance of Different Pathways of  $\text{CO}_2$  Fixation in Photosynthesis*) the reader is acquainted first with the method of  $\text{CO}_2$  fixation. High level answers are given to such questions as the ways of  $\text{CO}_2$  fixation (in  $\text{C}_3$ ,  $\text{C}_4$ , CAM species), the mechanism of  $\text{CO}_2$  concentration, or the special  $\text{CO}_2$  fixation of aquatic plants. With the knowledge of these basic concepts, the second part of the chapter discloses the ecophysiological aspects of mechanisms depending on the environmental conditions and becoming adapted to them. Chapter 16 (*Modelling of Photosynthetic Response to Environmental Conditions*) further deepens our knowledge of the subject, formulating these questions in mathematical models and giving an account of the most recent research results available. The model covers the biophysical, biochemical and physiological processes within the cell, the changes taking place in the organs and in the whole of the plant, and even some references to plant stands are found in this chapter. In Chapters 17 and 18 (*Regulation of Water Use in Relation to Carbon Gain in Higher Plants, Plant Life Forms and Their Carbon, Water and Nutrient Relations*) the ecological aspects are emphasized again. After a description of the "short-term regulation" and "long-term regulation" of the water regime, the properties of ecologically different plant types and the causes and physiological backgrounds of the differences are made known. The complexity of the question is indicated by the fact that beside the two environmental factors discussed so far ( $\text{CO}_2$  and water) the interaction of the nutrient conditions is also incidentally mentioned.

The indices of authors, plant names and subjects completing the book are well constructed, and assist the reader's orientation. This is necessary, since the knowledge imparted is extremely wide, and the book contains many concepts that are scarcely known or only recently introduced. Thus, volume 12B is a worthy continuation of the series started in 1981, and we look forward to the

appearance of Volumes C and D, which we hope, will not take long. In our opinion the series is equally useful for those engaged in plant physiology and ecology, and — in a certain respect — even for those interested in plant genetics and cultivation. They can profit by the material of the book in their everyday work.

J. BERNÁTH

ARMAND MAGGENTI: *General Nematology*. Springer-Verlag, New York—Heidelberg—Berlin, 1981, 372 p., 135 figs

Nematology and Helminthology, the two fields of science dealing with Nematodes, have diverged in several respects so far from one another that there is hardly any author who could undertake the task of summing up the most recent developments in both special lines.

Good books recently published on certain special fields of either Nematology or Helminthology are not at all few. Doubtless, however, there is a shortage of such up-to-date manuals as A. Maggenti's *General Nematology*.

This nicely produced, richly illustrated book of 372 pages is divided into 9 chapters.

Chapter 1 gives a brief historical survey of Nematology and Helminthology, primarily in their human parasitological aspects.

Chapter 2 deals with the morphology and taxonomy of Nematodes and of the phyla of helminths taxonomically closest to them (*Rotatoria*, *Gastrotricha*, *Kinorhyncha*, *Nematomorpha*). It should be noted that, while the author agrees in regarding the above "groups of animals" as the closest relatives of the Nematodes, the classification of these "groups" as animal phyla is far from being so unanimous.

Similarly, the authors differ in opinion concerning the taxonomic partition (phylum or class) of the nematodes and the further taxonomic division of their free-living parasitic forms.

In Chapter 3 to 5 the author discusses the morphological characteristics of *Nematodes*, taking into consideration most recent results of research.

Chapter 3 deals with the structure of the cuticle and secretory system of *Nematodes*, and with the process of moulting, which is an important phase in the ontogeny of *Nematodes*. Due emphasis is laid in the chapter on the new scientific results related to the structure, chemical composition and function of the cuticle. It is a well-known fact that the cuticular formations of *Nematodes* belong to the most important taxonomic characters, and the cuticle itself is a fundamental ele-

ment of the integrity of the worms. No matter what the strategy of *Nematode* control, the knowledge of the structure and properties of the cuticle is indispensable in its elaboration.

Chapter 4 discusses the internal morphology. It is here that the musculature, digestive and nervous systems of *Nematodes* are described.

In Chapter 5, as well as describing their genital systems, the spermatogenesis, oogenesis, and the embryonal and postembryonal development of the worms are dealt with.

Further chapters of the book give an excellent survey of the major types of *Nematode* parasites in plants, invertebrates and vertebrates.

Chapter 6 discusses the plant-parasitic species according to the classes set up by the author (*Adenophora* and *Secernentes*). The species *Longidorus*, *Xiphinema*, *Paratrichodorus* and *Trichodorus*, belonging to the first class, play an important role in the transmission of viruses in various cultivated plants. *Nematodes* of the second class redivided into parasites of underground and those of aboveground plant parts — in accordance with their two ways of development —, and are discussed through a number of suggestive examples.

Chapter 7 supplies information about several types of *Nematode* parasites among invertebrate animals.

Chapter 8 deals with the *Nematode* parasites of *Vertebrates*. The different forms of parasitism are described, the evolution of the parasitic *Nematodes* of *Vertebrates*, the best-known *Nematode* species are presented through examples taken from different taxonomic groups.

In Chapter 9 the author gives a taxonomic survey of the *Nematodes* and a characterization of the major taxonomic categories.

The book concludes with a bibliography and a subject index.

This book obviously will not satisfy those interested in special details of Nematology, but it was not intended as such. It may be recommended with the author's lines: "Hopefully this book will intrigue teachers, students, nematologists, plant pathologists, parasitologists, and zoologists. Each will approach the book from their own level of needs; some will read it superficially, some will delve into its speculations, and all, I hope, will learn to appreciate the science itself."

F. MÉSZÁROS



ARNOLD FINCK: *Fertilizers and Fertilization*. Introduction and practical guide to crop fertilization. Verlag Chemie. Weinheim. Deerfield Beach, Florida. Basel, 1982

The author, Dr. Arnold Finck, is professor at the Institute for Plant Nutrition and Soil Science of the University of Kiel (German Federal Republic). His book was originally published in German, similarly under the editorship of Verlag Chemie, in 1979, with the title "Dünger und Düngung: Grundlagen und Anleitung zur Düngung der Kulturpflanzen". The preface of the German edition, also translated into English, begins with the following sentence: "Why a new book on 'Fertilizers and Fertilization'?" The question is reasonable, since there are numerous comprehensive works on fertilization and fertilizers in the relevant literature. Professor Finck's answer to his own question is that the book had to be written because the books on fertilization problems published so far are either practical guides or else theoretical introductions; and today — some 100 years after the actual beginning of fertilization — it is possible and necessary to combine these two aspects. At the time of intensive farm management, it is not enough merely to observe some simple measures or general prescriptions, since at best these can only produce medium yields. Precise and comprehensive fertilization is required to satisfy the ever-increasing quantitative and qualitative demands for produce.

The author mentions that his book is based on university lectures of a period longer than twenty years, as well as on his innumerable discussions with expert colleagues in the inexhaustible topic of fertilization, both in his country and abroad. From the beginning he wrote his book for both local and international use. Fertilization is a question of world-wide importance, and many of its theoretical and practical aspects show similarities even under different ecological and economic conditions. In an intensive agriculture the purpose of fertilization is everywhere the same: to ensure optimum nutrients supply for crops often grown on insufficiently fertile soils under unfavourable climatic conditions, in order to obtain large yields. The concept of this book, although based on production conditions of the temperate zone, also takes into consideration modifying circumstances of the dry and the wet tropical climates. The international value of the work is enhanced by the fact that the major types of fertilizers applied in Europe are in similar use all over the world.

The main chapters of the book are:

- Introduction to fertilization
- General comments on fertilizers

- History of fertilization
- Mineral single-nutrient fertilizers: N-, P-, K-, Mg-, Ca- and S-fertilizers
- Micronutrient fertilizers: Fe-, Mn-, Zn-, Cu-, B-, Mo-micronutrients, and combinations thereof
- Multiple-nutrient fertilizer with major nutrients
- Combined macro- and micronutrient fertilizers
- Fertilizers with other nutrients: Na-, Cl-, Si-, Al-, Co-, CO<sub>2</sub>-fertilizers
- Fertilizers for soil improvement and general growth support: lime fertilizers, fertilizers for soil acidification, fertilizers for improving soil structure and texture, organic fertilizers, horticultural nutrient substrates, growth regulators, soil inoculants
- Optimal amounts of fertilizer: lime requirements, optimal contents of plants and soils, diagnosis of nutrient requirement, fertilizer recovery and nutrient removal, profitability of fertilization
- Special fertilization problems: methods of fertilizer application, influence of fertilization on the environment, fertilization as a function of soil type, fertilization as a function of the cropping system, fertilization under stress conditions, interpretation of results of field experiments
- Fertilization of agricultural crops: cereal crops, root- and tuber crops, oil crops and grain legumes, fibre plants and other industrial crops, fodder crops
- Fertilization in horticulture, forestry and special crops: vegetable and ornamental plants, fruit plants and vines, forest trees, stimulants, spices and medicinal plants, lower plants, e.g. algae, fungi, bacteria
- Fertilization and quality of vegetal food: concept and factors of quality, food quality as a function of the production system, fertilization system and food quality, food quality as function of nutrient supplies; fertilization, food quality and health of man and animal

Even the above outline provides sufficient information on the rich content of the book. In some 400 pages the author offers an excellent synthesis of the scientific results of modern agrochemistry and the practical experiences of up-to-date agriculture. Written in a clear didactic style, the book may be of great help primarily in university studies of agrochemistry and fertilization, though practical agriculturists can also make good use of it. The suggestive figure, and the synopses — emphasized by typographical means — which summarize the chapters, make it easy to survey the book and understand its content. The author displays ingenious handling of the possibilities inherent in typography to benefit expressiveness.



Most of the 307 works listed in the bibliography come — quite understandably — from German-speaking countries, but major English sources are also included in the list of references. It is regrettable — though to some extent natural because of the isolated nature of our language — that not a single Hungarian author is referred to in the bibliography.

The practicality of the book is facilitated and its text-book character emphasized by the list of units of measurement (SI!), the explanation of abbreviations, the tabular form of symbols and relative atomic masses of agrochemically important elements, the conversion of nutrients given in oxides into elementary forms — or vice versa —, and the definitions of chemical terms in the appendix.

Altogether, it can be said that Professor Finck's book has enriched agrochemical literature, and text-book literature in particular, with a truly valuable work. The highly representative English edition will certainly help to promote the book all over the world. As for us, we should welcome the Hungarian edition.

J. PECZNIK

E. BRESLER, B. L. MCNEAL, D. L. CARTER: *Saline and sodic soils. Principles-Dynamics-Modelling. Advanced Series in Agricultural Sciences 10.* Springer-Verlag, Berlin—Heidelberg—New York, 1982

Interest in salt-affected soils has recently increased all over the world. The reason is, on the one hand, that the development of soil science and many other natural sciences has opened up new possibilities to acquire a knowledge of salinization and sodification as a process, and to characterize the saline and alkali soils. On the other hand, the wide introduction of irrigation and the more intensive character of agricultural production demand increased attention to danger of soil salinization and alkalization. These causes may partly explain the fact that for some-years many books have been published on one problem or another of salt affected soils. Among them there is a great variety, from purely theoretical works to practical guides and manuals.

The book of Bresler, McNeal and Carter published by Springer-Verlag as Vol. 10 of Advanced Series in Agricultural Sciences belongs to the theoretical literature, as shown by the sub-title (Principles-Dynamics-Modelling), too.

Although the preface to the 236 page book promises both characterizations and description of alkali areas as well as informa-

tion about their utilization, the book deals with the physics and physico-chemistry of alkalization with a fundamentally theoretical approach.

The first part of the book, with the title "Diagnosis and Peculiarities", begins in a traditional way by discussing the origins of salts that cause salinization. Attention is paid to processes of salt accumulation that occur in nature, such as the salt content of sea-water getting into soils, the translocation of surface salt accumulation, or salt accumulation in the course of weathering. Salt accumulation processes caused by human activities are also discussed in this part of the book, although only briefly compared to the former ones.

Closely connected with the questions of salt accumulation, the quality of irrigation water is described together with the chemistry of soil solutions. The comparison and evaluation of methods used for measuring the salt contents of solutions, as well as their physico-chemical aspects are highly valuable parts of this chapter. The authors apply the analysis method elaborated by the US Salinity Labor, and work further on the basis of this measuring system. As a completion of this part the quantity and quality of harmful ions in the water or soil solution are briefly revived. Subsequently in this chapter, the physical properties of water occurring in the soil are characterized by the author, in the course of which the modelling of water potential, water retention and the system of soil-water-salts is described.

Further on, the interaction of precipitation and irrigation water with the soil is described, with emphasis laid on the ion exchange processes and the changes of minerals.

In describing the surface processes taking place in alkali soils, the authors briefly discuss the dominating clay minerals and the colloidal processes in the soil. In this context the effect of salts on the hydraulic parameters of the soil is also dealt with. The various models and equations of ion exchange are described on nearly 15 pages. Then the subject of salt solutions and precipitating, respectively, is treated, with special regard to the behaviour of carbonates.

In the final chapter of the first part, the diagnostical parameters of salinization and alkalization are summarized, for saline and sodic separately. This section is very short; it is a reference to the known parameters and their limiting values rather than a detailed analysis.

The second part, which deals with the transport and distribution of salts, sums up the different theories and equations of hydrodynamics; within this scope, it describes the effect of diffusion and convection on salt

transport, as well as the miscible displacement of salts. It is here that the authors discuss the chromatography of salt migration processes taking place in the soil and also show its theoretical background and methods.

Modelling of the migration of salts is described in detail, and the principles and ways of application of mathematical and numerical models are made known. This scope of subject forms one of the most elaborate parts of the book, perhaps closest to the authors' special line. This is suggested by the size of this section; it runs to nearly 50 pages, almost a quarter of the entire volume.

In connection with modelling, numerous cases are given by the authors. Among them an important place is occupied by the model of salt movement applied on various soils. Also, special models are described here for the interaction of soil and solution, and for the salt migration and dynamics of soil covered by plants.

The calculated and measured data are compared in many cases and in most models show good approximation. In this chapter the questions of soil heterogeneity related to the distribution and spread of solutions are also dealt with. Models are set for solution movements of various geometry to characterize the conditions produced by the heterogeneity of soils. In elaborating these models, attention is paid to the depth of the root zone. The models provide a good means for characterizing the probable concentrations of salt solutions at different depths of the soil profile. The sub-chapter on modelling offers a good start for those who study the theoretical questions of salt movement in the soil.

In the third part of the book, under the title "Management", the authors sum up all the questions that they consider important concerning the utilization of saline and sodic soils.

They begin with salt tolerance, then discuss both the osmotic and the specific ion effects. They speak of the sensitivity of plants to salt solutions in agreement with other literary data, and set up categories for plants suitable for production at a given salt concentration (sensitive, moderately sensitive, moderately tolerant, etc.). Detailed tables show the sensitivity of crops to salts and the prospective reduction of yield at given salt concentrations.

Besides the salt concentration, special attention is paid to the harmful effect caused by the exchangeable sodium percentage (ESP) on various crop groups in the case of low salt concentrations. Further, emphasis is laid in this chapter on the damage done by boron (B) to plants, provided that the quantity of this element in the irrigation water exceeds a certain limit value. Such

irrigation waters are frequently encountered in dry areas of the United States and in other arid regions.

This chapter also contains information on the quality of irrigation water. As to the quality and applicability of irrigation water, the authors apply the principles and methods of the US Salinity Labor as a whole, and give the same values as found in the manuals of that laboratory. They do the same in connection with the drainage and leaching requirement of soils.

The salt balance of soils is treated very briefly; it is a pity that here — like everywhere in the book — no reference is made to important results obtained in this regard within the Soviet Union and some other non-English speaking countries.

The same applies to the sub-chapter on the amelioration of saline and sodic soils, which gives a short review of the best known methods. The sub-chapter closes with a description of the economic model of soil amelioration and the technological model of irrigation. Finally, on hardly more than one page the authors mention special ways of utilization.

The book contains interesting and genuine material; it is to be regretted that some theoretical and practical works and source-books, though closely connected the subject in question, are not mentioned either in the text or in the list of references.

I. SZABOLCS

*Nucleic Acids and Proteins in Plants. II. Structure, Biochemistry and Physiology of Nucleic Acids.* B. Parthier and D. Boulter (eds). Springer-Verlag, Berlin—Heidelberg—New York, 1982, pp. 774

The 2nd volume of "Nucleic Acids and Proteins in Plants", vol. 14B of the New Series of "Encyclopedia of Plant Physiology", contains 18 chapters embracing all important areas of nucleic acid research in plants. The chapter "Nuclear chromatin" by W. Nagl deals with the chemistry of chromatin, the structure and function of nucleosomes, and chromosome structure. Clearly, in spite of a vast body of information available, we are far from understanding the structure of chromosomes and the exact role and function of chromatin. R. B. Flavell provides a chapter on "Chromosomal DNA sequences and their organization". Various methods of genome analysis are described, including renaturation kinetics, equilibrium centrifugation and the use of restriction endonucleases. Several instructive examples of genome organization are discussed. A lucid presentation of



"DNA replication and the cell cycle" is given by J. A. Bryant. Much attention is paid to the experimental systems suitable for the study of these problems. The replication of DNA in plant cells apparently follows the pattern studied in some more detail in animals. More work is, however, necessary to establish some specific features characteristic for plants. The next chapter, "DNA endoreplication and differential replication", by W. Nagl, stresses the significance of somatic polyploidization, a process which occurs much more widely in plants than previously thought. "RNA polymerase and regulation of transcription" is summarized in great detail by R. Wollgiehn. The message of the chapter is that more work is needed on plant RNA polymerases and, at this time, the regulation of transcription in plants is very poorly understood, despite the more than 250 papers referred to! A somewhat unusual title, "RNA sequences" (by T. A. Dyer), covers the established sequences of plant tRNAs, rRNAs and whatever was known about plant mRNAs at the time of writing. A comparison was made between the plant RNA sequences and the comparable RNA sequences in *Escherichia coli* and mammals. The next chapter by D. Grierson: "RNA processing and other post-transcriptional modifications", covers mostly what is known about rRNA, especially its cytoplasmic variants. Processing of tRNA is only suspected to take place in plants and clear proof is not yet available. Progress in the isolation and characterization of plant mRNAs is, however, rapid. A critical survey of "Ribonucleases and ribonucleic acid breakdown" is given by G. L. Farkas. Development in this field slowed down recently. The newly described RNases are poorly characterized and the role of the plant nuclei remains, in general, obscure. The review of C. Wasternack on the "Metabolism of pyrimidines and purines" deals primarily with the synthesis, interconversions and degradation of purine and pyrimidine nucleotides in relation to nucleic acid synthesis. The "Structure of plant viral genomes" is discussed by L. Hirth. The first part of this review pertains to the organization and the expression of cauliflower mosaic virus, which has a DNA genome. The second part of the review is devoted to the plant RNA viruses, and discusses primarily very new data in relation to the expression *in vitro* and *in vivo* of eukaryotic genes. A chapter on the "Translation of plant virus RNS's" by L. van Vloten-Doting and L. Neeleman devotes special attention to the possible functions of the structures of plant virus RNAs, and to the common modes of expression of the genetic information employed by different viruses. H. L. Sanger offers an extremely up-to-date

chapter on the "Biology, structure, functions and possible origin of viroids", in which the problems of host range, symptom expression, cytopathic effects, primary and secondary structures of viroids, linear and circular molecules, different viroid "species" and the replication of viroids are amply discussed. A review on "The Ti-plasmids of *Agrobacterium tumefaciens*", a bacterium which represents a natural system of genetic engineering in plants, is provided by J. Schell. Clearly, this is the single, most exciting system which offers perspectives of practical application, in addition to the wealth of data in the field of fundamental research which are already available. "The organization and expression of plastid genomes" is dealt with by H. J. Bohnert, E. J. Crouse and J. M. Schmitt. This chapter summarizes recent data on the physicochemical and structural aspects of plastid DNA, and physical maps of plastid DNA, as well as the transcription of the plastid genome. The application of recent techniques of molecular biology to plastid biochemistry is discussed in detail. Logically enough, this chapter is followed by another, written by W. Bottomley and H. J. Bohnert, on "The biosynthesis of chloroplast proteins". A main point emphasized is that the chloroplasts' synthetic machinery is less independent from the other compartments of the plant cell than previously believed. This idea is based partly on the "Use of mutants in the study of chloroplast biogenesis", a chapter written by K. W. Henningsen and B. M. Stummann. Although the subject matter of nucleio-chloroplastic interrelationship has partly been covered in earlier sections, due to the excellent experimental system offered by the giant alga, *Acetabularia*, this problem is dealt with in a separate review, "Interrelationship between chloroplasts and the nucleio-cytosol compartment in *Acetabularia*" by H. G. Schweiger. There is an extremely useful article by G. Gall on the "Use (and misuse) of inhibitors in gene expression". This is a field in which a high number of unjustified conclusions have been drawn and it was timely to draw attention to the pitfalls of indiscriminate use of inhibitors.

Like the previous volumes of the Encyclopedia, this one maintains the high quality of information that will serve as a standard source for plant physiologists and biochemists for many years to come.

G. L. FARKAS



NICKELL, L. G.: *Plant Growth Regulators—Agricultural Uses*. Springer-Verlag, Berlin—Heidelberg—New York, 1982. 173 p.

Plant growth regulators form an important group of the pesticides, since the growth and development of plants can be influenced with these compounds in the desired direction and extent. The agricultural and horticultural use of plant growth regulators has been found successful in many places, and owing to the larger yields obtainable with them, their role will increase in the future. All this is clearly stated in Louis G. Nickell's 1982 book "Plant growth regulators — Agricultural uses", which for this very reason, because of its topicality and encouraging intention, is welcomed by us.

The author of the book — Vice President of Research and Development at the Velsicol Chemical Corporation, Chicago, Chairman of the Plant Growth Regulator Working Group, and the Treasurer of the American Society of Plant Physiologists — discusses the effects of growth regulators on various plant functions.

According to the *Preface* the aim of the book is: a) to point out the enormous progress made in the last decades in the use of plant growth regulators, b) to make a survey of the relevant literature, c) to sum up the researcher's personal knowledge of this subject in both theory and practice up to the middle of 1980. The author pays, accordingly, little attention to the basic sciences that serve as a background, and to the action mechanism of the various growth regulators. In spite of this, each chapter contains a wide bibliography of a large number of summarizing works by authors who know the special aspects of plant growth regulators particularly well. The emphasis lies always upon the results achieved by now with a many-sided use of plant growth regulators.

Detailed instructions on using the different growth regulators for various purposes are deliberately avoided. In most cases the concentration or quantity required for the successful use of the compounds are not even stressed, since they greatly vary with species and variety, age of plant, environmental factors and geographical situation. On the rate of application, the reader can get detailed information from the bibliography. The screening methods that serve for the determination and evaluation of the activity of growth regulators are not dealt with either, owing to their high specificity, and because the author believes that the reader can also get a clear picture of these methods from the literature cited.

A discussion of plant growth regulators can be divided in a number of ways: by crop,

by chemical group, by physiological process and by miscellaneous classifications. In this book the growth regulators are discussed primarily from the standpoint of plant processes. By using this approach, it is easier to call the reader's attention to a) the practical and approved uses of plant growth regulators, and b) the stage of development of those chemicals as still under preliminary or advanced evaluation. In some chapters the division by plant process is not clear-cut because of the paucity of available practical information, which makes it necessary to combine two or more related topics.

The content of the book is divided into 24 chapters. The first of them is the *Introduction* which gives short definitions for the endogenous hormones that control the growth of plants, and for the non-hormone-like synthetic growth regulators from theoretical and practical points of view, and a brief characterization of the five groups of plant hormones. The *Introduction* also describes — extremely briefly — the mode of action of plant hormones, then surveys the 50-year history of their discovery and application in practice. Mention is made of the important change in the practical utilization of growth regulators after the slow development between 1940 and 1960. In the 1970's many new active ingredients were produced and new methods of use elaborated. It is a good thing that in these days the time between scientific discoveries and practical application is becoming shorter and shorter. As to the future, the rapid increase of world population makes it imperative to redouble the food production by the end of the century. The author is convinced that in this programme an important role will be played by the growth regulators, because their use offers almost boundless possibilities for the quantitative and qualitative yield improvement of many crops. According to the projections, these compounds will be produced in the coming years at a faster rate than other pesticides, becoming thereby one of the most rapidly expanding sectors of the agricultural chemical industry.

The discussion of the practical application of plant growth regulators is divided in the book in the following chapters:

2. Rooting and plant propagation (2 pages, 50 citations)
3. Germination and dormancy (2 pages, 37 citations)
4. Flowering (7 pages, 82 citations)
5. Gametocides (4 pages, 57 citations)
6. Abscission (9 pages, 114 citations)
7. Fruit set and development (4 pages, 33 citations)
8. Plant and organ size (13 pages, 95 citations)

9. Axillary buds (2 pages, 21 citations)
10. Chemical pruning (2 pages, 31 citations)
11. Plant shape (1 page, 10 citations)
12. Tillering (1 page, 10 citations)
13. Resistance to, and control of, insects and diseases (4 pages, 50 citations)
14. Overcoming environmental stress (4 pages, 53 citations)
15. Mineral uptake (1 page, 5 citations)
16. Plant composition (3 pages, 40 citations)
17. Metabolic effects, ripening and yield increases (26 pages, 322 citations)
18. Modification sexual expression (2 pages, 24 citations)
19. Senescence (2 pages, 51 citations)
20. Desiccation (6 pages, 107 citations)
21. Protection against herbicide damage (4 pages, 46 citations)
22. Increase of herbicide absorption and translocation (1 page, 3 citations)
23. Toxicology, environmental and human safety (3 pages, 41 citations).

Each chapter begins with a short theoretical part followed by a concise description of practical applications. In chapters of major importance the compounds to be used are put in tables.

On the first pages of chapter 24, the *Summary*, the high importance of plant growth regulators in agricultural production is re-emphasized, successes possibly achieved in the future are outlined and the significance of basic and applied research stressed. The author's hopes concerning the future are reflected in the following quotation: "By 2000 A.D., I predict that plant growth regulators, primarily as yield enhancers, secondarily as quality improves and production process facilitators, will become widespread for world crop production".

On the following 16 pages of the *Summary* a table is found in which all growth regulating compounds mentioned in the book are listed in alphabetic order. The table gives the chemical name, common name, designation or code, trade name, the plant growth regulating activity, other activity, and the chapter and location. This systematic summing up to the nearly 200 compounds is very useful for those engaged in studies on the subject.

The *Literature* with references to 1201 publications is perhaps the most valuable part of the book, since this work was intended to be a review of the available literature rather than a customary manual. Most of the publications listed were written between 1960 and 1980, mainly by American authors, though there are among them a considerable number of works from other countries. It is a pity that many of these publications are hardly available for us.

The book ends with indices of authors and subjects.

Owing to the objective of the book, the text is extremely concise, and is restricted always to the most necessary information. Thus it was possible to summarize almost completely all aspects of the use of growth regulators in such a small space. The 29 illustrations are good quality photos of which more would have been welcomed in the book.

The appearance of L. G. Nickell's book is really timely, as the last important work in the subject was published ten years ago (R. J. Weaver: *Plant growth substances in agriculture*, 1972). During the last ten years the greatest progress has been made in the use of plant growth regulators. The book is thus a stop-gap work in current professional literature, which encourages its readers to acquire a better knowledge of the plant growth regulators and widen their practical application.

M. VARGA

G. BALÁS, GY. SÁRINGER: *Kertészeti kártevők (Horticultural pests)*. Akadémiai Kiadó, Budapest, 1982

Géza Balás, the inspirer and first author of this book, had already published the results and experiences of his several-decade teaching and research work in 1963, then in an enlarged and revised edition in 1966, under the title: *Kertészeti növények állati kártevői (Animal pests of horticultural plants)*. The present bulky volume contributed to by notable co-authors was published under the editorship of the Akadémiai Kiadó. Its basis and backbone are provided by the two editions of the above-mentioned work.

The authors dedicate their work to Gusztáv Szelényi, doctor of biological sciences, titular professor and retired head of a scientific section, who has opened a new epoch and established a school in the Hungarian plant protection entomology. As an epigraph they cite an almost forgotten Latin saying: "In hoc libro mortui vivant et muti magistri loquantur!" (May the dead revive and the mute Masters speak in this book!). The pair of reputable authors have done their work in this spirit.

The Preface was written by András Somos, professor, academician, and vice-president of the Hungarian Academy of Sciences. In the introduction the authors inform the reader about the theoretical considerations and professional experiences on which the work is based, and about the experts and colleagues who directly or indirectly assisted them in their work.

The book consists of four large parts (Generals, Ecology, Plant protection and Particulars), and completed by an appendix



of uniformization of Hungarian insect names, bibliography, and index of names and subjects.

The value of the work is considerably increased by the fact that within the main section of Ecology a part of the autecology and the chapter of gradology were written by Tibor Jermy, academician, while the synecology by Professor Gusztáv Szelényi; in the detailed part, the species of the order Physopoda are discussed by Gábor Jenser, candidate in agricultural sciences, head of a scientific section.

The authors introduce their subject with an extremely varied and colourful historical survey from ancient times to the present, followed by the history of research and education of horticultural zoology in Hungary, and a chapter on the most prominent Hungarian representatives of horticultural zoology. The authors took good care to acquaint the reader with the activity of the most famous representatives of the profession between 1755 and 1799: János Földi, Ottó Herman, Gusztáv Emich, József Pavlavszy, Géza Horváth, Károly Sajó, Albert Szaniszló, Sándor Lovassy, József Jablonowski, Károly Schilberszky, Gábor Bakó, István Pásztor, Gyula Kadocsa, Jenő Györffy, Márton Aczél, Gábor Reichart.

In the chapter on harmful and useful animals these questionable and often debated concepts are clearly defined. Under the superscription "Increase in the number and importance of harmful species" the reader can form a true picture of the questions indicated. The following, seemingly shorter chapters (Grouping of animals by feeding, The way of feeding) duly connect the previous parts with the subsequent ones. The chapter: *The Concept and Practical Importance of the Picture of Damage* is excellent both logically and didactically. It is a pity that the chapter does not include illustrations of the more typical damages.

The second large section: Ecology, occupying nearly 100 pages, is a highly valuable part of the work. The sub-chapters, written with a highly up-to-date view of the subject, contain even the most recent research results. It is obvious that the authors (Jermy, Sáringer, Szelényi) are internationally recognised ecologists. The chapter clearly expresses that a successful, up-to-date, environmentally protecting pest control can no longer be carried out without ecological knowledge.

The third large section — Plant protection — discusses the plant protection in general, the prevention, the methods of pest control, and the integrated plant protection. Within the latter, the authors present the results of recent investigations into the feeding inhibitors, the sexual attractants (sex-feromons) and the tracer feromons. To com-

plete the chapter, the development of protective means up to the appearance of sprayers is discussed.

In the second large, detailed section the reader is informed about the necessity of animal systemization, the concept of species and the origin of species. The description of species begins with the Nemathelminthes and ends with the Vertebrata, in accordance with their respective taxonomic places. Full particulars are given of the host plants, damages, ethology and economic importance of more than 500 (!) species, and of the directives of control. After the Hungarian and Latin name of each pest, the full name of the author describing the species as well as the Hungarian and Latin synonyms of the pest are given. Excellent illustrations make the contents still more valuable.

One of the great values of this section is that wherever possible the authors quote from old notes and letters asking for advice (from 1664!). In this way the reader learns that pests thought to be new in our time may have already caused — and did cause — problems in the 17th century.

At the end of the fourth large section, an alphabetic survey of pests according to their feed plants and the plant parts damaged is presented. The general pests and the species feeding on a great variety of plants are mentioned by the authors with those plants which the respective pests like the best. This mode of systemization is particularly useful to the practical experts.

In the Appendix the authors deal with the theoretical questions of making the Hungarian insect names uniform. Their intention deserves credit, as species are often mentioned in the literature by six to ten different Hungarian names; and conversely, the same Hungarian name is sometimes used to designate 3 different species. With a view to choosing the names properly, the first author of the book (G. Balás) sums up his remarkable proposals in 8 points.

The Bibliography, as previously mentioned, is extremely rich, and even the 2500 data of bibliography were included after a thorough selection. Not only the student of universities and colleges preparing their diploma theses, but also teachers and research workers, can make good use of it.

The book closes with indices of author's names and subjects, respectively.

As for the directives of control discussed with the individual pests, the most recent results are mixed with earlier experiences, particularly in the field of plant protectives. It is thus clear from this work too, how quickly the pesticides recommendable for control become obsolete. The authors therefore properly recommend active ingredients



instead of trade-names in most cases. Should this cause any uncertainty, the reader will find further data in the publication "Növényvédő szerek és műtrágyák" (Pesticides and fertilizers) edited every year — as suggested by the authors themselves.

For the photos J. Bodor editor, professor L. Móczár (doctor of biological sciences), the late G. Reichart, doctor of agricultural sciences, scientific consultant, L. Szalay-Marzsó, candidate in agricultural sciences, senior member of a research institute and L. Migend, research worker deserve credit. The figures are the work of E. Pataki, candidate in biological sciences, emeritus assistant professor and K. Biró, engineer designer, department member.

All in all, the authors have completed an enormous work, of which nothing similar on horticultural pests has been published so far either in Hungary or abroad. If only for the large number of illustrations, research workers and experts of plant protection as well as owners of home gardens — not to mention the students of universities and colleges — can make good use of this book.

Of the authors G. Balás, candidate in agricultural sciences, was for years professor and head of the Department of Entomology at the College of Horti- and Viticulture (now University of Horticulture) and reputed teacher and researcher of entomology for decades. Gy. Sáringer, doctor of agricultural sciences, is one of the founders of experimental insect ecology in Hungary, an internationally recognised researcher, scientific consultant earlier to the Research Institute for Plant Protection, at present to the Plant Protection Institute of the Keszthely University of Agricultural Sciences.

S. BOGNÁR

J. BERNÁTH—T. TISCHNER—A. ÁBRÁNYI:  
*Plant Environment and its Control*. Akadémiai Kiadó, Budapest, 1982. 241 pages

The work of the three authors a biologist, an electroengineer and a physico-mathematician, is the first to sum up in the Hungarian language the biological effects of natural and artificial environment, the possibilities of control and the planning and evaluation methods of experiments. Beside their international experiences, each of the authors gives the results of his own work. Up-to-date investigations into the plant environment require the joint work of representatives of the related sciences, for which this handbook is a good example. References to nearly two hundred Hungarian and foreign literary works provide further guidance to those interested in more details. The 101 text

figures, 25 tables and 21 photos make the handbook suggestive. The six tables in the appendix contain useful data.

Initiated by Went three decades ago, the importance of artificially controlled plant environment researches was soon recognized in Hungary, and biological research work which met the requirements of the technico-scientific revolution was already started at the end of the fifties. On the basis of international experiences, a phytotron was built at Martonvásár, and a climatron at Budakalász. Apart from these two large internationally known and acknowledged establishment, growth chambers are used in many biological and agricultural research sites, and the purchase of such facilities is scheduled for further locations. This handbook may thus excite wide interest.

The introduction recalls the initiators of phytotron methodics in Hungary — Prof. Imre Horváth and Sándor Rajki academician. Each of the authors has been doing phytotron work for more than ten years and plays an important role in the propagation of Hungarian phytotron methods.

The first chapter discusses the biological importance of natural and controlled environment, and describes the natural model of the environmental factors, the relation of plant to environment, and the model of controlled environment.

The history, the biological and technical background of the artificial control of plant environment are described in the second chapter. Information on the development of phytotron methods in Hungary and on the prospective trend of development is also given here.

The third, largest chapter deals with controlled climatic factors. In sub-chapters on light, temperature and air, the reader is acquainted with the natural conditions of the respective factors and their effects on plants, as well as with the technical possibilities of control and the methods of measurement.

The natural conditions of radiation, the laws of radiation which are of basic importance for plant life, and everything known about radiation reaching the Earth are summed up by Andor Ábrányi, who began his career as an agrometeorologist.

The effect of light on plants is explained as manifested in photosynthesis, photoregulation, photomorphism and phototropism.

The methods of light control are described by Tibor Tischner, the first Hungarian phytotron engineer. The subject matter is grouped by spectral composition, radiation intensity, period of illumination, light sources, incandescent lamps, low pressure gas dis-

charge fluorescent tubes and high pressure gas discharge tubes. The figures and photos clarify the contents of the sub-chapters. In the tables, the data of various light-sources are shown.

In the sub-chapter on radiation measurement the deficiencies of illumination measurement (in lux unit) — the widest used unit even today — are discussed. To make the experiments repeatable the data expressed in lux unit have to be completed with those of the lighting apparatus. The radiation technical units of measurement are given according to the SI system. The basic principles of measuring the radiated energy, and the description of the most up-to-date instruments available are demonstrated by numerical data, diagrams and photos.

Under the title, *Natural Temperature Conditions* essential information is given by the physicist author on the heat regime of the Earth, heat conductivity and temperature conditions of plant and soil. Changes in air and soil temperature over time and space are also described.

The biologist author writes about the factors influencing the heat balance of the plant, thermal adaption of plants, effect of temperature on photosynthesis and dry matter production, and the temperature relations of growth and development. In the same sub-chapter we can read about the effect of extreme temperatures.

The control of temperature, the major structural elements of climatic systems and the cooling media are so summarized by the engineer author as to show the biologist, who uses or intends to use the phytotron methodology, the possibilities offered by the present level of technics.

In the following section the basic principles of temperature measurement and the major types of measuring instruments are discussed. When measuring the air temperature, protection from radiation must be provided in a controlled environment as well. The fundamental problems of leaf temperature measurement are also shown.

Of the components of air, carbon dioxide and vapour are the most important for the life of plant which are summarized by the physicist author. Their natural conditions, further, the characteristics of the vapour content, the absolute humidity, the specific humidity, the vapour pressure, the relative humidity, the saturation deficiency and the dew point are treated in the same chapter.

The composition and motion of air is of basic importance for the life process of plants. They are dealt with from various aspects, in accordance with the authors' respective special fields: the role of carbon dioxide, oxygen, air pollution, vapour con-

tent and motion of air, and the possibilities of controlling and of measuring them. The infrared gas analyser, the most generally used measuring apparatus, the ways of avoiding the errors and the methods of measuring are described.

The next sub-shapter sets forth the methods of air humidity measurement, and groups the hygrometers together with their deficiencies according to their working principles. For the use of the most frequently applied hair hygrometer, practical advice is given.

The last sub-shapter briefly sums up the principles of air flow measurement. This section is too short, and those interested in the subject can get complete information only from the literary references.

In the following main chapter the biological role and possibilities of control of the soil, and of water supply and nutrient status within it, are discussed.

In the phytotron methodology, the term growth medium is used instead of soil, because it is not only in soil that plants can be grown under controlled conditions. The adjustment of the physical factors of the growth medium ensures optimum conditions for the root system.

The water supply and its control, the translocation of water, the water discharge, the water regime of plants, and the measurement of the water content in the growth medium may cause many problems in the phytotron technique. Even now the exact water content of the growth medium can only be determined by thermogravimetry. Apparatus working on various principles are also mentioned, though their accuracy is insufficient for research work.

The sub-chapter, *The Chemical Properties of the Growth Medium and Their Control*, deals with the ecological role of the soil pH, the nutrient supplying capacity of soil and growth medium, the mineral nutrient uptake by plants and the control of nutrient supply.

In the sub-chapter, *Biotic Factors of the Growth Medium*, the relationships of plant — microorganism and of higher plant — plant are summarized.

In the sub-chapter, *The Artificial Media of Controlled Plant Growing and the Culture Pots*, the physical constants of the most frequently used growth media and their mixtures are given in a tabulated form. Useful practical information is supplied as to the material and size of culture pots.

The fifth chapter surveys the currently used conditioned plant growing units and phytotrons.

In the sixth chapter, the physicist author summarizes the methods of climatic programme planning and climate simulation model construction. The latter means that



those climatic conditions as most frequent in the open are provided for the given plant. This system has been used and has proven itself in the Martonvásár phytotron for a decade.

In the chapter on experiment planning and -evaluation the physico-mathematician author's experiences are summed up and the literature of the subject reviewed. In artificial plant growing spaces, much fewer plants can be studied than in the field, therefore the errors must be minimized. The chapter discusses the ways to achieve this goal. By a comparative trial of field- and phytotron populations, the author demonstrates the variations of plants raised in the phytotron and in the field. As an example the arrangement, data survey and analysis of variance of a trifactorial phytotron experiment are presented.

The book ends with the chapter, *Operation of Plant Growing Climatic Apparatus*. Apart from an abundance of literary data, ten-year experiences of the electric engineer author are summarized in it, and advice is given on how to choose the apparatus. The reliability of the apparatus is examined by mathematical methods. The necessary conditions of putting it into operation are described. As to the organization work required for the operation of the phytotron the author modestly says that no prescription can be given for universal utilization. This subchapter, the summary of ten years of experience at Martonvásár, however, provides advice, utilizable by every phytotron operator.

Finally, we must mention the six tables of the appendix which contain a bulk of useful information.

The editor Gyula Pál deserves credit. For the style and structure of the handbook, and the arrangement of the figures, tables and photos.

J. PLETZER

P. FRIEDRICH: *Supramolecular Enzyme Organization. Quaternary Structure and Beyond*. pp. 1-300. Pergamon Press, Oxford and Akadémiai Kiadó, Budapest, 1984.

This book is devoted primarily to a highly up-to-date area of enzymology: the structural and functional aspects of enzyme complexes, the association of "soluble" enzymes with each other and a variety of cellular structures. It is certainly not a new standard textbook of enzymology, of which quite a few are available. The book is rather a monography which reflects the main field of interest of the author and his co-workers

during the past few years. However, in order to make the message of the book accessible for the average biochemists, some fundamental principles of enzyme structure and function are also described. Still, the basic interest of the author is neither the primary, secondary, tertiary nor even quaternary structure of enzymes. The focus of attention of the text is extended to those problems of enzymology which are *beyond* the quaternary structure.

Three introductory chapters on "The hierarchy of enzyme structures", "Chemistry of protein associations" and "Intramolecular organization: the quaternary structure of enzymes" provide sound basis for the discussion of three major problems: "Intermolecular organization: multienzyme systems", "Multienzyme complexes and conjugates", "Association of enzymes with cellular structures". The author stresses the fact that structural organization *between* different enzymes (e.g. enzyme juxtaposition) is much more common than previously thought and the study of *links*, or *communication*, between enzymes has for a long time been neglected. Excellent examples for structural organization are described. It is pointed out that multienzyme *complexes* catalyse a number of metabolic processes. These complexes are stable and can be isolated both as entire complexes and as individual enzyme molecules. In multienzyme *conjugates* the individual enzymes catalysing the individual enzymatic steps are covalently bound proteins. Even typically "soluble" enzymes (e.g. those of glycolysis may interact. The author shows how this interaction facilitates the "orderly" processes taking place in nonstructured or slightly structured media. Metabolic compartmentation is also discussed as a closely related phenomenon. A number of examples are also listed in which the enzymes are structure-bound (e.g. bound to membranes) and it is explained how this attachment alters their properties. In some cases it even makes the proper physiological functioning of the enzymes possible.

The book is written in a very good, clear style, and is a pleasure to read. It entertains, in addition to the description of facts, a host of new and provocative ideas which will raise interest in the subject among many readers. The scope is enormous; 1351 works are cited! It is regrettable that the publishing procedure was so slow. (The review of literature was completed in 1981 and the book appeared in the second half of 1984.) However, quality of the photographs, and even of some drawings, is not up to the standard of the text.

The book is highly recommended not only to the enzymologists but also to all those biochemists who wish to learn of the enzym-



ology of the 1980s and the directions it might follow in the next decades.

G. L. FARKAS

R. HUNT: *Plant Growth Curves. The functional Approach to Plant Growth Analysis*. Edward Arnold Limited, London 1982; pp. 248, ISBN 0 7131 2844 5

The experimenter who growth plants makes sequential observations on the plants, the subject of investigations. Parallel to the observations he performs various kinds of measurement, so the series of measuring carried out at different times shows the course of development of the individual plant parts or of the whole plant. The description of growth does not require higher level mathematical analyses, as long as measuring is confined to one and the same specimen; but, for example, for the determination of the dry matter production, the plant material must be dried, which means that at the successive dates of sampling, data of different plants are recorded. In this case, beside the genotypic variation of the plant population, further sampling, phenotypic, etc. mistakes are added to the errors of the experiment. To describe the growth of plants, mathematical functions are thus reasonably applied here. A similar problem appears when, under the influence of various environments, species or varieties show different trends of growth — in which case it is much simpler to compare them by the parameters of the growth functions.

Despite the fact that the book was written by a biologist for the attention of other biologists, its subject matter is strongly mathematical and statistical. This must not, however, discourage anybody from reading the book, as a standard knowledge of algebra, and of differential and integral calculus is sufficient for understanding. Those who wish to get acquainted with the different methods in more detail can choose among nearly 600 literary references.

The mathematical analysis of growth is encountered in many similar publications of the Edward Arnold Limited, for example in Causton, D. R.: *A Biologist's Mathematics* (1977), Clarke, G. M.: *Statistics and Experimental Design* (1980), as well as in another book by the author of the present work, Hunt, R.: *Plant Growth Analysis* (Studies in Biology, No. 96, 1978). The same Edward Arnold Ltd. published a book on the biometric bases of the subject, *The Biometry of Plant Growth* by Causton, D. R. and Venus, J. C. (1981).

Roderick Hunt's book is divided in eight main chapters. The introduction acquaints

the reader with the growth data of maize published by Kreuzler et al. in 1879, which forms the basic data for presentation of the regression methods discussed in the subsequent chapters. It is here also that the system of notation used in the book, the SI units of plant parameters, and the computing support required to carry out the curve fitting, in most cases no other than a programmable pocket calculator, is described.

Chapter 2 discusses the theoretical bases of the classical parameters of growth dynamics (absolute and relative growth rate, net assimilation rate, leaf area ratio, leaf area index, etc.) for individual plants and populations alike, as well as their interrelations. At the end of the chapter a comprehensive table of the classical parameters of plant growth is found. Besides their names and abbreviations, this also provides their definitions and units, as well as the pages on which their particulars can be found.

The third chapter deals with the theoretical basis of functional approaches; emphasizing the role of empirical models in connection with curve fitting to plant growths data. In this chapter, useful formulae are given by the author for the relation of the classical plant growth parameters to those obtained by the functional approach in the case of both the original- and the log-transformed basic data.

The subsequent four chapters discuss the practical methods of the functional approach and its function types, from the curve drawn free-hand for the basic data, to the most complicated splined regression that requires a computer. The researcher, naturally, has to decide whether the application of one or another type of regression is necessary, and if so, which of the methods should be used. These questions are answered in Chapter 4. The same chapter tells about the statistical background of the curve fitting, emphasizing three important prerequisites which exist for regression analysis: firstly that an independent variable ( $x$ ) should be measured without, or virtually without error; secondly, that the distributions of replicated  $Y$  values at each  $X$  should be normal; thirdly, that the variance of these subpopulations of  $Y$  should not change in magnitude with  $X$ . The first condition is not difficult to satisfy, since in the case of plant growth the  $x$  variable is generally that of time, which can be measured with much greater accuracy than the data of the plant. As to the fulfilment of the second and third conditions, a number of publications are cited by the author in which various transformations are applied to the problem's solution.

Chapter 5 deals with the first, second, third and high order polynomials. The de-

scription of each type is followed by a table of literary reviews, which contains the names of authors, the species examined, the frequency of sampling, the primary and derived data, and short comments. The application of functions to the growth data of maize, given in the introduction of the book, is carried out for the different order polynomials. Then, from their figures, we get a highly suggestive picture of the advantages or disadvantages of these individual methods. The author subsequently discusses publications on the application of stepwise regression polynomials.

The subject of Chapter 6 is similar, various asymptotic functions are mutually compared. A separate sub-chapter deals with the monomolecular, logistic, Gompertz', Richards' and other similar types of functions. After Richards', a generalized asymptotic function formula is presented, which together with the annexed table describes all possible asymptotic growths.

The seventh chapter discusses special functional approaches. These methods may be less known in practice, although they are based on the polynomials that, in turn, can be widely used. In the case of segmented regression, the measuring data series is divided into segments, and the functional approach will of several slightly overlapping functions. A logically improved variation of this is the running re-fit, or in other words the moving, continuous regression. As to its type, it resembles the technique of moving averages. It is followed by the splined regression, a chain of separate polynomial functions which join in the so-called "nodes". The latter, while requiring a computer, exceeds the former methods concerning its results. At the end of the chapter the problems of analysing the time series are described, in addition to some types of regression that differ from those so far used.

In the last chapter of the book the author touches upon some problems arising in practice, such as too many data for classical analysis, or too few for functional approach.

All in all, we can say that Hunt's book can be of use not only for those studying the growth of plants but also for other biological experimenters. Its wording is simple, easy to follow, and the numerous literary references offer much help to those who wish to go deeper into the subject.

A. ÁBRÁNYI

*The amazement of a passionate gardener* (Ed.: D. SURÁNYI), Magvető, Budapest, 1982.

This thick volume of a series of the Magyar Hírmondó (Hungarian Courier) offers

much pleasure to the reader. The sound account of the epilogue traces Hungarian horticulture back to an Eurasian past. The preface begins with a prehistoric survey, in which the author seems to have adopted the conception of Péter Hajdu and István Fodor that the original home of the Hungarian people was Western Siberia, and that later the "Ugrian home" was transferred to this side of the Urals. Alas, these opinions are highly questionable or even improbable, and the latest works are not always the most reliable ones. It is not from a single territory that the prehistory of the Hungarian people began; it encompassed a large part of Western Eurasia. The author fortunately does not build upon the "original home" idea, nor upon the conjectures about the centuries preceding Árpád's conquest of Hungary; and the phrase, "the crisis of the pastoral society", that sounds well but is far from true, is missing from his terminology. The recent prehistorical investigations quite rightly take into account the animal farming and agricultural activities in the environment of the areas of possible interest. As a result it has become generally known that the agricultural activity of the Hungarian people was shaped over thousands of years before the conquest of Hungary, and only became richer in the Carpathian basin. The influence of monastic orders and of the Slavs in the bordering areas is unquestionable, but only their influence; their teaching was not new. Dezső Surányi himself held to the right proportions when discussing these questions.

We advise the readers to begin by reading the epilogue because the learned editor offers an excellent characterization of sources and ages in it. We do the same in now reviewing this book.

The scarce sources before the conquest of Hungary are followed by documents which tell us mainly of the viticulture of Hungary, though they also mention kitchen-gardens, farm-yards and orchards. Much can be learnt here about early Hungarian fruit- and vegetable production. The collection covers equally the gardens of landlords and serfs. Recent authors' summarizing works inserted among the documents and contemporary records offer the possibility of a comprehensive survey. At the dawn of the new age after the Middle Ages, theoretical works and textbooks by Hungarian authors (Péter Melius Juhász, János Apáczai Csere, János Lippai, and accounts of the Turkish times) already appeared, and the book cites ample selected passages from them. It is with these suggestive descriptions that we arrive at the New Age, and from works of those times, selections are found in similar abundance throughout the book. The splendid selections



generally end with material from the nineteenth century.

I should like to contribute a few remarks for a possible new edition. First of all, let us have the contribution of the archeologist in the preface. What might he find in this brilliant collection that could help him in his work? Unexpectedly, an apparently interesting fact presents itself with which we are going to deal at some length.

In graves of the migration period, especially in the late Avar—Onogur cemeteries, eggs placed beside the deceased are quite often found. For a long time we accepted the belief that the eggs in the graves of girls and young women were the symbols of fertility. At the Department of Archeology the University burial with eggs has been the subject of two dissertations. Without drawing the conclusions and dropping the theory of fertility-enchancement, both authors wrote about eggs placed not only in graves of young women, but sometimes in those of children and even of men. On the occasion of a recent scientific examination of egg finds, the latter turned out to be hatched eggs. By these two observations, the fertility theory lost its credibility and a new explanation had to be found. A possibility was offered in the *Calendarium* by János Lippai (1661). Namely, for the month of March it reads: "Shells of eggs from which chicks have hatched this month should be collected and preserved; they are very useful as medicine against gout". So, chance has offered the possibility of a new interpretation; namely, the eggshell was placed as a medicine beside the person who had died of some disease. This is only a conjecture, but worth considerations.

The text contains frequent references to the curative power of plants. An example from János Lippai's book "*Veteményeskert*" (Kitchen-garden) (1664): "This fruit" — cucumber — "is considered by the doctors as not very healthy, because its juice, especially when much of it is consumed makes people feel very sick, and may cause inflammatory diarrhoea difficult to cure. Nevertheless, with the seeds boiled and the brew drunk the stagnant urine can be removed. It abates the inflammation of the kidney, eases the fit of ague, the severe inflammation, heat and thirst." Similar health-giving qualities of plants are endlessly listed. So far, when dealing with the floral patterns of the Middle Ages and subsequent times, we have not paid much attention to the fact that the flowers and fruits were embroidered, painted or glazed on objects not only for their beauty, but also for their role in preventing or curing diseases (a few more words about this are given below). Perhaps it was for the same reason that flowers were planted at the front

of houses and in the gardens; the housewife kept a real pharmacy at hand, or dried plants in the loft or cupboard. This book encourages the ethnographers to carry on further investigations into the remains of the Herbarium of the Middle Ages and into the medicinal knowledge popularly preserved. Let me recall my childhood, when I clearly remember that the only medicine of my grandmother at Székelyszáldobos was the juice of sauerkraut. She drank it when she had a headache, when she cut her hand, or when any kind of misfortune happened to her.

István Kovács, the unforgettable master of Transylvanian Hungarian archeology, called my attention to the fact that the palmette leaves of the first Hungarian settlers represent succulent plants which suggest the nearness of the sandy steppe. Investigations have also begun on the plant representations of late Avar—Onogur bronze finds (grape-vine varieties) and from the character of the grape-vine varieties in the Nagyszentmiklós find, Miklós Füzes-Frech' drew the conclusion that in the environment of the find there had been vine arbour cultivation.

Even these few remarks suggest that, with archeological discoveries, Hungarian horticulture can be traced back to a period much earlier than the time of the first documents. The results of analyses of seeds and pollens, from the time of the original Hungarian settlers at Felgyő, will soon be published, whereby we shall be able to gain knowledge of the horticulture of those days.

Altogether, we must have the courage to investigate periods preceding the time of written texts and, although we cannot establish facts, at least set up well-founded theories.

Take, for example the matter of tulips. The book acquaints us with a study by Sándor Takáts, the great researcher of the Turkish age in Hungary on this subject. He was a master of archival research and the written word, and in his opinion the tulip was introduced in Hungary by Turkish gardeners in the 16th century, which contemporary documents and the accounts of travellers suggest. Yet, if we consider the descriptions of flowering steppes by those who had been there, it becomes evident that, in the eyes of people living among them, they were the materialization of beauty. When their girls are praised, the image of the steppe with its spring tulips emerges again and again. This picture is reflected in the carpets covering the inside of their *jurtas*, and also in the Mohammedan mosques, with their brilliant glazed tiles, in which the blooming steppe turns into a paradise. It is in this Arabian Nights' atmosphere that our girls' flower names, which mainly after the crusades spread all over the



West, take significance and in this atmosphere the flower songs become meaningful even if the texts have been adjusted to the symbols of Christianity. Julia, the pretty girls (a figure from a Hungarian poem) goes to pick wild flowers in the field, and a bunch of wild flowers was placed on the dead body of a girl at the time of the first Hungarian settlers (dissertation by Csanád Bálint). It is thus possible that the documents at hand only trace back the events and facts to the sixteenth century, which leaves us at the time of the Turkish occupation of Hungarian, but the presentiment of an earlier reality takes us back to the tulips blooming on the steppes.

As another problem to be investigated, I mentioned to my students that the floral decorations on earthenware, for example, might also be brought into connection with the role of the represented plants as spices for foods and drinks. So far we have taken the glazed or painted bunches of flowers for the merely aesthetic manifestations, but it is possible that they suggest more than that.

Besides their usefulness as medicines, the flowers have been given a symbolical meaning, primarily in the field of love. This is a wonderful mixture of natural phenomena and colours. Initial steps have already been taken towards investigations on this subject.

The value of the book lies not only in its data; it also gives the reader further food for thought. The sight of flowers and fruits becomes imbued with spirit for us, just as in the Herbaria of the Middle Ages.

Beside these mystical aspects, we must not forget that the flowers are exhilarating to the sight and their existence makes one feel pure. Our tables are often decorated with bunches of flowers and our walls with flower-pieces. It is these that bring the freshness of nature into our homes.

All in all, this brilliant book brings pleasure not only to the garden enthusiasts, but may be a permanent companion to those as well who long for beauty and harmony.

GY. LÁSZLÓ

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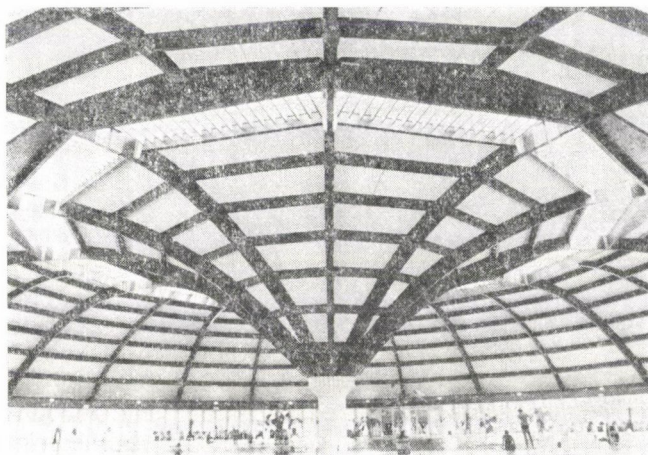
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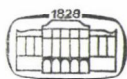
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33. VORTRAGSTAGUNG UND MITGLIEDERVERSAMMLUNG  
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GESELLSCHAFT FÜR ARZNEIPFLANZENFORSCHUNG



33rd ANNUAL CONGRESS  
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1 | LIPID AND VOLATILE CONSTITUENTS FROM CELL CULTURES  
OF ALYSSUM MINIMUM WILLD.

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Allysum minimum Willd. is a member of the family Cruciferae, well known for the presence of glucosinolates. The plant is distributed through Europe and the Middle East and has diverse medical usage, its major application in Iran being as a febrifuge. No previous work has been carried out on the plant, however the presence of isomeric methyl-thio-isothiocyanates has been noted in seeds of A. chondrogynum and A. troodi.<sup>1</sup>

— Volatile isothiocyanates were extracted from seed and dried plant material after autolysis, along with free fatty acids and hydrocarbons. The major isothiocyanate was found to be 3-butenyl, (365µg/g in dried plant) but smaller amounts of allyl isothiocyanate were also detected. Four nitriles and an epithiobutane derivative were also identified and estimated along with nineteen n-alkanes and five fatty acids. All compounds were identified using capillary GC-MS plus RRT measurements. Callus and suspension cultures were established on Murashige and Skoog's medium with a variety of hormonal supplementations and again 3-butenyl isothiocyanate was found in callus cultures which had been supplemented with 0.2-1.0ppm 2,4-D or NAA plus 0.1 or 0.5ppm kinetin, but could not be detected in suspension cultures. C10-C28 alkanes and five fatty acids occurring in seeds and dried plants were also found in callus and suspension cells. The major seed and whole plant fatty acids were in decreasing order; oleic (C18:1) palmitic (C16), and erucic (C22:1), whereas the major fatty acid of callus and suspensions was palmitic acid.

1. X. Hasapis, A.J. Macleod and M. Moreau, Phytochem. 20 (1981) 2355.

2 | PHOTOSYNTHETIC PIGMENTS IN DIFFERENTIATING DIGITALIS  
LANATA SUSPENSION CULTURE

L. Mannonen<sup>1</sup>, R. Hannus<sup>2</sup>, P. Hynninen<sup>3</sup>, V. Kauppinen<sup>1</sup> and L. Björk<sup>4</sup>

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Digitalis lanata suspension cultures produce cardenolides only after the cell aggregates have differentiated into greening embryoid-like structures. A special cultivation scheme has been developed for enhancing the differentiation (1). In this, cells grown in the dark with high auxin concentration are gradually accustomed to grow in light with decreased auxin and higher cytokinin concentrations.

We have been interested in finding an instrumental indication for the commencement of the differentiated growth of the cardenolide-producing suspension culture of D. lanata. To this end we undertook to follow the appearance of some photosynthetic pigments and simultaneous structural differentiation at the multicellular and cellular level.

The pigments were extracted in acetone, concentrated and separated by TLC. The components were identified by comparing their R<sub>f</sub>-values, and fluorescence to standards of  $\beta$ -carotene, chlorophyll a and b, luteine and pheophylline a. The morphological differentiation of D. lanata was followed both visually and microscopically.

(1) C. Kuberski, H. Scheibner, C. Steup, B. Diettrich and M. Luckner, Phytochemistry **23** (1984) 1407-1412.

### 3 | DER CHITOSAN-EINFLUSS AUF DIE SYNTHESELEISTUNGEN VON NICOTIANA TABACUM ZELLKULTUREN

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Chitosan, ein lineares  $\beta$ -1,4-Polyglucosamin, kann als Bestandteil pflanzenpathogener, pilzlicher Zellwände vorkommen. Eine Reihe von Befunden weist auf seine Rolle bei der Beziehung Wirtspflanze-Parasit hin: Chitosan kann als Elicitor in befallenen Gewebsteilen eine Phytoalexin-Akkumulation mit fungistatischer Wirksamkeit auslösen, aber auch andere Abwehrmechanismen sowie eine erhöhte Synthese von Phenolen, Terpenoiden, Polyinen und Alkaloiden bewirken (1). Da pflanzliche Zellkulturen nutzbare Sekundärstoffwechselprodukte meist nicht oder nur in geringen Mengen produzieren, erhebt sich die Frage nach der Beeinflussbarkeit solcher Stoffwechselwege durch Elicitoren.

Als Modellsystem dienten Suspensionskulturen von *Nicotiana tabacum*, die durch Variation des Kulturmediums (Phytohormone) Nicotin in mit der Ausgangspflanze vergleichbaren Mengen produzierten (Nicotinbestimmung (2)). Mit zunehmendem Kulturalter erhöhte sich die Nicotin-Akkumulation (% TG). Chitosan induzierte die Biosynthese von Phenolen und die Lignifizierung der Zellwände, erhöhte die Plasmalemma-Permeabilität (reduzierte Fluorescein-Akkumulation) und wahrscheinlich über verstärkten Ca-Influx (3) auch die Callose-Biosynthese (Anilinblau-Färbung und Methylierungsanalyse). Die Nicotin-Synthese chitosinbehandelter Zellen war jedoch - ohne Wachstumshemmung - deutlich erniedrigt. Fütterungsversuche mit  $^{14}$ C-Asparaginsäure erwiesen, daß unter Chitosaneinfluß der Einbau von  $^{14}$ C-Markierung in Nicotin zugunsten einer erhöhten Protein-Biosynthese reduziert war.

- (1) B. Wolters and U. Eilert, DAZ, 14 (1983) 659
- (2) M.T. Pinol, J. Palazon and M. Serrano, Plant Sci. Lett., 35 (1984) 219
- (3) H. Köhle, W. Jeblick, F. Poten, W. Blaschek and H. Kauss, Plant Physiol., 77 (1985) 544.



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Werden Pflanzen unter Sauerstoffabschluß kultiviert, so sind viele Arten zur anaeroben Glycolyse fähig, wobei Pyruvat häufig in Ethanol umgewandelt wird. In pflanzlichen Zellsuspensionskulturen treten hypoxische Bedingungen relativ leicht, besonders am Ende einer Kulturphase auf, wenn die Zelldichte hoch ist. In dieser Phase produzieren Lupinenzellkulturen aktiv Ethanol. Als maximale Ethanolkonzentration konnte 1% Ethanol im Zellkulturmedium gemessen werden. Ein ähnlicher Effekt wurde in Zellsuspensionskulturen von 8 weiteren Pflanzenarten festgestellt (1). Der Einfluß unterschiedlicher Sauerstoffkonzentrationen auf das Zellwachstum und die Ethanolproduktion wird dargestellt. Unter Sauerstoffabschluß, direkt nach dem Überimpfen von Lupinenzellen in frisches Medium, konnte eine Induktion der Alkoholdehydrogenase und parallel dazu der Ethanolbildung nachgewiesen werden. Es wird diskutiert, in wieweit die in Zellsuspensionskulturen auftretenden hypoxischen Bedingungen auf den Differenzierungsgrad der Zellen und damit die Bildung pflanzlicher Sekundärstoffe haben können.

(1) M. Wink, J. Plant Physiol., 120 (1985) 287-293.

INDUCTION OF ANTHRAQUINONE BIOSYNTHESIS BY BIOTIC  
ELICITORS IN CELL SUSPENSION CULTURES OF CINCHONA  
LEDGERIANA MOENS

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In previous studies (1,2) we reported on the isolation and identification of anthraquinones from callus cultures of Cinchona ledgeriana. In cell suspension cultures of the same species anthraquinones are also formed. Although the genus Cinchona has been subject to extensive chemical studies, only one paper seems to have been published indicating that the genus Cinchona might contain anthraquinones (3). The reason for the occurrence of relatively large amounts of anthraquinones in tissue cultures, the occurrence of which has still not been definitely proved in the intact plant, might be that the callus tissue (in fact a sort of wound tissue) forms anthraquinones for its protection against infections. Because the anthraquinones isolated from C. ledgeriana tissue cultures show a clear antimicrobial activity, the hypothesis was developed that these anthraquinones act as phytoalexins. In the present study we report on the induction of the anthraquinone biosynthesis in cell suspension cultures of C. ledgeriana by addition of a sterilized mycelium filtrate of Phytophthora cinnamomi, a fungus known to be pathogenic to Cinchona species. Upon addition of the filtrate mentioned to suspension cultures of C. ledgeriana, the anthraquinone content of the culture increased sevenfold, as compared with a non-treated culture. It thus seems that the anthraquinones mentioned may indeed act as phytoalexins.

1. Mulder-Krieger, Th., Verpoorte, R., de Water, A., van Gessel, M., van Oeveren, B.C.J.A. and Baerheim Svendsen, A., Planta Med. 46, 19-24 (1982).
2. Wijnsma, R., Verpoorte, R., Mulder-Krieger, Th. and Baerheim Svendsen, A., Phytochemistry 23, 2307-2311 (1984).
3. Covello, M., Schettino, O., La Rotonde, M.I. and Forgione, P., Bell. Soc. Ital. Biol. Specim. 46, 500-503 (1970).

6 | GROWTH AND SECONDARY METABOLISM IN *CINCHONA* CELL CULTURES  
AFTER ADDITION OF INDOLE ALKALOID PRECURSORS

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Cell suspension cultures of *Cinchona* were grown in Gamborg B5 medium to which tryptophan, tryptamine or secologanin was added. Tryptophan added at T=0 inhibited growth nearly completely at all concentrations tested (0.1 - 10 mM) with no significant effect on the quinoline alkaloid production and slight decrease of anthraquinone production.

Secologanin caused some decrease in dry-weight yield of the cultures, anthraquinone production increased slightly with increasing concentration of the iridoid. Lower concentrations of added tryptamine resulted in slight increase of dry weight. At higher levels, however, a decrease was observed. Addition of tryptophan in the exponential growth phase resulted in a rapid metabolism of this precursor, whereas growth is not inhibited at the lower concentration levels (0.1 mM). Analysis of the alkaloid extracts of the tryptophan feeding experiments showed the presence of  $\beta$ -carboline as the major component.



7

Axenische Massenzucht von Mikroalgen in  
Mittelkulturen (20 - 300 l) und in einer  
Pilot-Anlage (3000 l)

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Ziel der Untersuchungen ist die Verwendung von Mikroalgen für Zwecke der Ernährung und für die Gewinnung von pharmazeutisch relevanten Inhaltsstoffen.

Mit Hilfe einer neuartigen Technologie (T-förmiger kombinierter Belüftungs- und Saugrührer mit niedriger Drehzahl; 1-3 U/min) werden fädige Blaualgen (Cyanobakterien) in geschlossenen Becken (20-3000 l; mit Innenbelichtung) unter axenischen Bedingungen gezüchtet. Die angewendete Technologie eignet sich für alle Arten von Mikroorganismen und für die Zellkulturen. Sie ermöglicht vollständige Suspendierung und optimale O<sub>2</sub>-Versorgung. Wegen der äußerst geringen Scherkräfte bleiben die Zellen unzerstört. Das Abernten erfolgt nach Sedimentation auf rationelle Weise (unter erheblicher Volumeneinsparung) über Absaugkammern in den Seitenarmen des Rührers.

Die Untersuchung von Blaualgen zeigt, daß man bei diesen Organismen durch Variation verschiedener Umweltfaktoren (Belüftung, Belichtung, Temperatur, Zusammensetzung der Nährlösung u.a.m.) sowohl die proteinreiche Biomasse als auch die Bildung bestimmter Algeninhaltsstoffe manipulieren kann, wie z. B. Lipide, Fettsäuren, Sterole und Carotinoide.

M. Piorreck, K.-H. Baasch und P. Pohl, *Phytochemistry* 23 (1984) 207

M. Piorreck und P. Pohl, *Phytochemistry* 23 (1984) 217

8 | COMPARATIVE STUDY OF THE ALKALOID CONTENT OF DIFFERENT  
STRAINS OF PSYCHOTRIA MANNII GROWING IN VITRO AND PLANT LEAVES

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Different Psychotria species (Rubiaceae) studied in our laboratory produce alkaloids of indolin type. The alcoholic extract obtained from in vitro culture of Psychotria mannii Hiern. revealed the probable presence of alkaloids (1). These preliminary results will be confirmed by HPLC analysis of both extracts obtained from some stable strains and the entire plant.

METHODS

- a) Tissue culture  
P. mannii strains consist of either callus or cell suspensions, growing on Murashige and Skoog medium supplemented with different growth factors (2,4-D + KIN, ANA + KIN).
- b) Extraction and analysis  
Freeze-dried cell cultures and powdered leaves were extracted using the same procedure adopted for Psychotria in our laboratory (2).

RESULTS AND DISCUSSION

Four alkaloids were detected in the leaves powdered extract. The major one was detected in the callus of each strain as well as cell suspension of three weeks subculture. In addition, two of the studied strains are found to be able to produce alkaloids normally absent in the plant leaves. Both Psychotria mannii leaves and cell cultures seem to produce alkaloids of polyindoline type of more than five units in a contrast with those found in other Psychotria species.

- (1) C. Linder, A. Roth, R. Anton, Colloque consacré aux Plantes Médicinales, Angers (1983)
- (2) A. Roth, Thèse de 3e cycle, Université Louis Pasteur, Strasbourg (1984)

# INDUSTRIAL PRODUCTION OF SECONDARY METABOLITES FROM PLANT CELLS IN SUSPENSION CULTURE: A FEASIBILITY STUDY

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The aim of the research into plant cell biotechnology by the cooperating Universities of Leiden and Delft is the industrial production of secondary metabolites. To investigate the economics of this production, a feasibility study was done.

A process was designed for the production of 50 kg of an alkaloid X. The plant cells were assumed to have a specific growth rate of  $0.164 \text{ day}^{-1}$ , a yield of 0.5 kg biomass/kg sucrose, the alkaloid content of the cells was assumed to be 1% of dry weight at the end of the production phase. On the basis of literature data, fed-batch culture in air-lift fermentors was selected as the production system.

To attain the planned production, an inoculum was cultured in a  $0.1 \text{ m}^3$  fermentor for 2 weeks. The contents of this fermentor were transferred as inoculum to a  $1 \text{ m}^3$  fermentor. The contents of the  $1 \text{ m}^3$  fermentor were used alternately to inoculate two  $10 \text{ m}^3$  fermentors. The growth period in the  $10 \text{ m}^3$  fermentor was assumed to be 2 weeks and the alkaloid production period also 2 weeks. The equipment and the growth media were supposed to be steam-sterilized.

The price of the product was calculated to be approximately 5000 Dfl. per kg (1200 US\$), neglecting the costs of down stream processing and man power. The price was determined as: investment costs (70%), energy costs (15%), and medium costs (15%). To decrease this price a shorter production cycle to decrease the investment costs is evidently necessary. This can be achieved by higher growth and production rates of the cells, or by recycling the biomass in stead of sacrificing the cells.



10 | PRODUCTION OF INDOLE ALKALOIDS BY TISSUE CULTURES  
OF TABERNAEMONTANA SPECIES

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The production of indole alkaloids by a callus and a suspension culture of Tabernaemontana elegans was compared with the production by the intact plant. There were some striking differences. In the plant extract corynanthean alkaloids (vobasine, dregamine and tabernaemontanine) were the main components. In the callus culture the aspidospermatan alkaloid apparicine was the main component. Together with vobasine, apparicine forms the dimeric indole alkaloid monogagine, which was only isolated from the callus culture. From the whole plant extract only dimeric indole alkaloids of the coynanthean-iboga type were isolated. The suspension culture produced up to 0.015% (on fresh weight) alkaloid on a standard MS medium. The alkaloid mixture produced by the suspension culture contained at least 25 different alkaloids.

Suspension cultures of another species, Tabernaemontana divaricata, grow with doubling times of 60-75 h. They produce several monomeric indole alkaloids (e.g. apparicine, vobasine and tubotaiwine). Saccharose, nitrate and phosphate up-take from the medium by this suspension culture agrees well with the data published for Catharanthus roseus.

12] CARDENOLIDE FORMATION IN DIGITALIS LANATA PLANTS PROPAGATED  
VIA MERISTEM CULTURE

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High-yield Digitalis lanata plants are needed for the commercial production of cardiac glycosides. Until now, such plants have been obtained by inbreeding. The inbred lines are crossed and high-yield hybrids are selected which are then used for seed production for commercial plantation. Digitalis is a biannual, meaning that this process always has to be repeated. It would be of great advantage to be able to maintain and multiply those plants which are used for seed production. Common vegetative propagation with cuttings is not practicable, because only 20% form roots. A method for multiplying one parental plant a thousandfold in a relatively short time by using meristem culture has been described previously(1). If, however, one wants to use such plants for commercial seed production, one has to ensure their genetic identity, especially with respect to cardenolide formation.

In this paper we will present the analytical data of Digitalis plants propagated by meristem culture. Leaf samples were extracted with 70% ethanol according to the method described by Wichtl et al. (2), using  $\beta$ -methyldigitoxin as an internal standard. Cardenolides were analysed by high performance liquid chromatography (HPLC) using a Hewlett Packard 1084 B Liquid Chromatograph equipped with a stainless steel column (Nucleosil 5 C 18, Macherey & Nagel; length 25cm, diameter 4.6mm). Cardenolides were eluted with a non-linear gradient (solvent A: re-distilled acetonitrile (84%); solvent B: water): 30-42% A (35 min), 42-50% A (5 min), 50-60% A (15 min). Temperature conditions: oven 40°C, acetonitrile 30°C, water 50°C. Measuring wave length: 225nm, reference wavelength: 350nm.

Digitalis lanata plants were regenerated from long-term meristem cultures (5-19 months) and analysed after 7-17 months of growth in a greenhouse. All of the regenerated plants were very similar with regard to both their cardenolide pattern and lanatoside C content. Further investigations will clarify questions of biochemical stability in Digitalis lanata during long-term culture.

(1) Schöner, S., Reinhard, E.: *Planta medica*, 45, 155 (1982)

(2) Wichtl, M., Wichtl-Bleier, W., Mangkudidjojo, M.: *J. of Chromatography*, 247, 359-365 (1982)

A method has been developed of retaining the growth rate and biosynthetic capacity of a cell strain of Coleus blumei after freezing in LN.

Of several compounds(DMSO, glycerol, mannitol, proline, saccharose, sorbitol) tested for their cryoprotective action, sorbitol in high concentrations showed the most effect combined with the lowest toxicity for Coleus blumei cells, a strain established by the Nattermann & Cie. GmbH.

When added to the medium on the 4th day of a 7-day-cycle, sorbitol in 1M concentration gave the best results after an incubation time of 8-16h. Cells pretreated in this manner are partially dehydrated and need no further cryoprotection in the two-step freezing regime used. The cooling rate was 1°C/min down to -40°C. This temperature was held for 40 min to allow equilibration before plunging the probes into LN. Ending the controlled freezing process at this temperature yielded satisfactory results.

Best recovery was reached when the ampoules were taken from the LN and thawed within 70 sec in a water bath of 62°C.

Viability was tested immediately by FDA, regrowth was obtained on filter-paper discs over agar.

When callus growth was maintained, suspension cultures were easily reestablished and tested for their ability to still form rosmarinic acid(2).

The amount of rosmarinic acid formed by cells which had undergone a freeze-thaw cycle was in the same order of magnitude as that reached with untreated controls.

The procedure worked out, which utilises the protective and osmotic dehydration capacity of sorbitol(1,3), is a suitable protocol for the cryoconservation of Coleus blumei cell cultures in LN.

LN = liquid nitrogen(-196 °C)

- 1) T.H.H. Chen, K.K. Kartha, N.L. Leung, W.G.W. Kurz, K.B. Chatson, and F. Constabel (1984), Plant. Physiol. 75, 726
- 2) B. Ulbrich, W. Wiesner, and H. Arens(1985), Proc. der Tagung "Primary and Secondary Metabolism in Plant Cell Cultures." 6.-8.9.84 in Rauischholzhausen, Springer-Verlag, in press
- 3) G. Weber, E.J. Roth, H.G. Schweiger (1983), Pflanzenphys. 109, 29



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The root of dandelion, Taraxacum officinale Weber is said to have anti-rheumatic, cholagogue, laxative and diuretic actions<sup>1</sup>.

The therapeutic activity of dandelion has not been ascribed to a single known constituent. However, because of its world-wide reputation as a diuretic it was decided to investigate these properties and it was hoped to more accurately identify the agent(s) responsible for this activity.

Powdered dandelion root material was successively extracted with petroleum ether, chloroform and methanol. From the crude extracts, various fractions were chromatographically separated and further purified.  $\beta$ -amyrin and  $\beta$ -sitosterol were verified as being present.<sup>2</sup>

The crude extracts and some purified fractions were pharmacologically evaluated for diuretic activity.<sup>3</sup> Three of a total of five groups (3 mice per group) were orally dosed with 50 mg/kg of each extract suspended in 0.25% sodium carboxymethylcellulose (SCMC) in saline (0.9%), one group with an equivalent volume of the vehicle and the fifth group with frusemide, a known diuretic in a dose of 37.5 mg/kg. The urine from each group was collected over a period of 5 hours, volumes being measured after 1, 3 and 5 hours. Urine  $\text{Na}^+$  and  $\text{K}^+$  concentrations were determined after 5 hours by Atomic Absorption Spectrophotometry.

No consistent pattern of diuresis was obtained with the extracts of dandelion root, although two of the extracts showed statistically significant natriuretic and kaliuretic effects. This possible diuretic effect in mice must be considered along with the reported activity of Taraxacum officinale in rats.<sup>4</sup>

Tissue cultures were initiated from dandelion seeds and grown in callus and suspension cultures using a modified Linsmaier and Skoog medium. Extracts from callus and suspension cultures were found to contain the same range of compounds which were chromatographically similar to those present in whole root extracts.  $\beta$ -amyrin and  $\beta$ -sitosterol were identified as occurring in the tissue cultures. Extracts were not subjected to pharmacological screening.

1. British Herbal Pharmacopoeia, Part I, 1976, published by British Herbal Medicine Association.
2. Kasprzyk, Z., Grzelczak, Z. and Pyrek, J., Bulletin de l'Academie Polonaise des Sciences Cl. II. Vol. XIII, No. 11-12, (1965), 661-665.
3. Sim, M.F. & Hopcroft, R.H., J. Pharm. Pharmacol., 28, 609-612, (1976).
4. Rácz-Kotilla, E., Rácz, G. and Solomon Ana, Planta Medica 26, 212-217, (1974).

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Phytohormones influence the growth of plant cell cultures, as well as their morphological and biochemical differentiation (1). During prolonged cultivation, however, the cells may become independent of the auxins and cytokinins routinely supplied with the culture medium. This process is called habituation (2). Habituated cells may be more stable than non-habituated ones in terms of their capability to perform specific biochemical reactions, e.g. biotransformations.

Our aim was to establish and characterize habituated cell lines from Digitalis lanata callus. Cell strains differing in their ability to biotransform  $\beta$ -methyldigitoxin (3) were selected for hormone-autotrophic growth. The resulting four cell lines (K 10H - K 40H) were further characterized. The time courses of uptake and biotransformation of various cardiac glycosides (digitoxin,  $\beta$ -methyldigitoxin,  $\beta$ -methyldigoxin, and lanatoside A) have been established. The cardenolides were quantified by HPLC (Nucleosil C 18; non-linear gradient: acetonitrile-water). In addition we determined cell growth, protein content, and some enzyme activities ( $\alpha$ -mannosidase,  $\beta$ -glucosidase, acid phosphatase, malate dehydrogenase, and glucose 6-phosphate dehydrogenase). This strategy yielded "fingerprints" of all four habituated cell lines. These fingerprints are presented and discussed. In summary, they give a more precise view into the biotransformation reactions performed by cell cultures of Digitalis lanata, especially with regard to in-vitro studies necessary for characterizing the enzymes involved in these reactions.

In addition, we expect to use these well characterized cell lines in further investigations into the stability (during long term culture, fermentation, and cryopreservation) and manipulation (with phytohormones, light, antimetabolites, etc.) of foxglove cell cultures.

- (1) Mantell, S.H. and H. Smith (1983) in: Plant biotechnology, pp 75 - 108. Eds: S.H. Mantell and H. Smith. Cambridge University press
- (2) Everett, N.P. (1981), J.Exp.Bot. 32, 171
- (3) Heins, M. (1978), doctoral thesis, Tübingen

16 | Alternative Bildung von Anthrachinonen und Lipochinonen  
in heterotrophen und photoautotrophen Zellsuspensions-  
kulturen von Morinda lucida Benth.

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Es wird die Regulation der Synthese von Lipochinonen und Anthra-  
chinonen an Zellkulturen von Morinda lucida untersucht.

#### Methoden

Die Suspensionskulturen wurden in Doppelstockkolben (1) oder in einem  
Fermenter (2) kultiviert. Die isolierten Lipochinone wurden IR-, MS-  
und UV-spektroskopisch charakterisiert. Die quantitative Bestimmung  
der Lipochinone und Anthrachinone erfolgte UV-spektroskopisch.

#### Ergebnisse

Photoheterotrophe und photoautotrophe Zellsuspensionskulturen wurden  
von Kalluskulturen von Morinda Lucida Benth. angelegt und charakteri-  
siert. Der Gehalt an Lipochinonen in photoautotrophen Kulturen ist  
vergleichbar mit dem in einem intakten Blatt. Anthrachinonglykoside,  
die in den Wurzeln von Morinda gefunden wurden, waren in der photo-  
heterotrophen Kultur nur in Spuren, in der photoautotrophen Kultur  
jedoch nicht vorhanden. Anthrachinonsynthese in großer Menge wurde  
beobachtet, wenn photoautotrophe und photoheterotrophe Kulturen ins  
Dunkle gebracht wurden und das Medium Saccarose enthielt. Die Induk-  
tion der Anthrachinonsynthese fiel mit einem raschen Verschwinden der  
Lipochinone zusammen. In den Suspensionskulturen fällt also Photo-  
autotrophie mit Lipochinonsynthese und Heterotrophie mit Anthrachi-  
nonbildung zusammen. Dies spiegelt die Verhältnisse in der intakten  
Pflanze wieder, wo Lipochinone in Chloroplasten vorliegen, während  
Anthrachinone in der Vakuole vorkommen.

#### Literatur

- (1) Hüsemann, W., Plohr, A. und Barz, W., Protoplasma 100, 101 (1979)
- (2) Hüsemann, W., Protoplasma 113, 214 (1982)



17 | HPLC SEPARATION AND QUANTITATIVE DETERMINATION OF  
ALOE-EMODIN, EMODIN, CHRYSOPHANOL AND PHYSCION IN  
PLANT CELL CULTURES OF RHAMNUS FRANGULA AND RH. PURSHIANA

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Plant cell cultures can accumulate compositions of anthracene derivatives which are different from those in the intact plants. In plant cell cultures of Rhamnus frangula and Rh. purshiana, developed and investigated in our laboratory, 1,8-dihydroxyanthracene derivatives mainly occur as anthrones and dianthrones, besides the anthraquinone forms. These oxygenated anthracene derivatives were found to accumulate as such and as glycosides (1). Reference samples of such derivatives are rare and sometimes difficult to obtain.

In order to study the production of oxygenated anthracene derivatives by plant cell cultures, a procedure was elaborated by which originally present free and glycosidic bound anthracene derivatives were converted into the corresponding anthraquinone aglycones (2).

HPLC separations based on gradient elution are less useful to perform routine quantitative determinations. For the separation and quantitative determination of the anthraquinone aglycones obtained from the cultures, an isocratic HPLC procedure is presented for good resolution of aloe-emodin, rhein, emodin, chrysophanol and physcion within a relatively short time (3).

(1) A.J.J. van den Berg and R.P. Labadie, *Planta Med.* 50 (1984) 459

(2) R. Kinget, Thesis, Catholic University of Leuven, 1966, 90

(3) A.J.J. van den Berg and R.P. Labadie, *J. Chromatogr.* 329 (1985)

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18 | UNTERSUCHUNGEN ZUR FRUCTOSAN-BIOYNTHESE IN  
ZELLKULTUREN VON SYMPHITUM OFFICINALE

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Als Vorversuche zu Untersuchungen zur Regulation des primären und sekundären Kohlenhydrat-Stoffwechsels in Zellkulturen wurden aus Blättern, Blattstielen/Sproß, Wurzeln und Fruchtknoten von Symphitum officinale Zellkulturen angelegt. Die Calli wurden auf verschiedenen Medien mit unterschiedlichen Phytohormonkombinationen und -konzentrationen unter verschiedenen Kulturbedingungen (Licht/Dunkel) gezogen (1). Nach mehreren Passagen unter den jeweiligen Kulturbedingungen wurden die Zellen und ein Teil der Kulturmedien wäßrig extrahiert. Nach Ausfällung der Polyphenole mit Pb-Acetat wurde der Extrakt konzentriert und papierchromatographisch auf Oligo- und Polysaccharide, insbesondere Fructosane, überprüft (2) und die Zuckersamensetzung GC quantitativ bestimmt (3).

Die aus Blättern angelegten Calli bilden Fructosane in größeren Mengen. Bei den aus Wurzeln, Blattstielen und aus Fruchtknoten isolierten Calli liegt der Gehalt an Fructosanen unter der Nachweisgrenze. Die Phytohormonkombination und -konzentration, sowie die Art des Nährbodens haben keinen direkten Einfluß auf die Fructosanbildung. Die Fructosanbiosynthese erfolgte lichtunabhängig. Alle Kulturen enthielten große Mengen an Polyphenolen, wobei im Dunkeln angezogene Kulturen besonders hohe Konzentrationen aufwiesen.

Weitere Versuch mit auf neuen Medien angezogenen Zellkulturen und mit Regeneraten aus Wurzeln und Sproß/Blattstiel-Calli sollen zeigen, ob die für die Fructosanbiosynthese verantwortlichen Enzyme nur in den Zellkompartimenten im Cytoplasma der Blätter aktiv sind, und daher nur in Zellkulturen aus Mesophyll eine Fructosanbiosynthese möglich ist.

(1) A.A. Abou-Mandour, *Planta Medica*, 46 (1982) 105

(2) C.S. Wise, R.J. Dimler, H.A. Davis, and C.E. Rist, *Anal. Chem.*, 27 (1955) 33

(3) L.T. Sennello, *J. Chromatogr.*, 56 (1971) 121

19 GENETICAL AND CHEMICAL STUDIES ON *CORYDALIS*  
CAVA L. TUBER TISSUE CULTURES

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In this investigation we aimed to follow the chromosomes variation in *Corydalis cava* L. tuber tissue cultures in correlation with alkaloid and polyphenol contents. Callus cultures grew in dark on Murashige Skoog (1961) solid medium containing IAA, 2,4-D, Kinetin, as growth regulators and glycine, tyrosine, phenylalanine amino acids as supplements.

For cytological studies three weeks old young tissues were fixed in Farmer solution and stained with the usual acetocarmine technique. Total alkaloid and polyphenol content were measured according to the method of Lőrincz and Szász (1961) and Swain and Hillis (1959) respectively.

Tissues showed large variation in their chromosome numbers from the diploid (16) to the polyploid from 54 chromosomes. The diploid cells in the tissue were ranged from 47.71 % to 61.30 %. This variation in the percentage of diploid cells depends on the type and concentration of growth regulator or supplement. The data indicated that the presence of Kin (1 mg/l) and IAA (2 mg/l) was the best concentration for the biosynthesis of both alkaloids and polyphenols. Absence of glycine from the MS medium decreased the total alkaloid content by 50 percent. The results revealed that the polyphenol content increased when tyrosine and phenylalanine were added to the MS medium (4 mg/l). Data showed good correlation between the high percentage of polyploid cells and high content of secondary plant products in the callus.

- (1) Lőrincz, Cs., Szász, K.: Acta Pharm. Hung. 3, 106 (1961)
- (2) Murashige, T., Skoog, F.: Physiol Plant 15, 473-479 (1962)
- (3) Swain, I., Hillis, W.E.J.: Sci.Ed.Agric. 10, 63-68 (1969)



21 | INVESTIGATIONS INTO THE STABILITY OF TABERSONINE  
PRODUCTION OF AMSONIA TAXA

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Several taxa of the Apocynaceae family, such as *Amsonia angustifolia* Ait., *A.tabernaemontana* Walt., *A.illustris* Woods. *Rhazya orientalis* A.DC. owing to the indole alkaloid /tabersonine/ content of their seed /1-2/ may serve as raw material for the semi-synthesis of compounds of high pharmaceutical activity /e.g.vinpocetine/.

It was the aim of investigations to establish the possible role of selection in increasing the alkaloid content and to determine the factors influencing the tabersonine content.

Seed samples of populations, sown in 1964 and propagated both vegetatively and generatively, were extracted with petroleum ether. Tabersonine content was determined photometrically, at 328 nm.

The variability in the alkaloid content of plant individuals within the populations was smaller than that of the samples taken from the different levels of the same individual /at the same time/, however it exceeded the values of the average of seed samples of populations /20 m<sup>2</sup>. Actually, the age of populations does not affect the tabersonine content. Yearly variations of ecological factors and soil types have moderate effect. In view of both these findings and the study of the progenies of plant individuals, the gradual selection of populations of higher tabersonine content seems to be possible. Thus, in 1984, the average content of *Amsonia* taxa was /0.74+0.12/%, while that of *A.angustifolia* populations increased to /0.82+0.18/%.

Data on the seed protein pattern /gained by PAGE/ as possible means in characterizing *Amsonia* and *Rhazya* taxa are also discussed.

/1/ B.Zsádon, L.Décsei, K.Otta, M.Szilasi, P.Kaposi

Acta Pharm.Hung. 44, 74 /1974/.

/2/ Jr.I.Máthé, MTA Biol.Oszt.Közl. 25, 427 /1982/

22 | OCIMUM SANCTUM L. AND OCIMUM CITRIODORUM VIS.:  
INCREASE IN PRODUCTION, CONTENT AND ANTIMICRO-  
BIAL ACTION OF ESSENTIAL OIL

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Ocimum sanctum L. was treated to increase considerably its production in herb material by cutting off the main stem during early stage of growth. Due to this procedure the content of essential oil was enlarged at the same time.

In general, synthesis of essential oil in Ocimum undergoes a distinct seasonal dependence.

The average amount of oil produced in fresh leaves was in the range of 0.07 to 0.09%, and 0.08 to 0.1% in the fresh flowers. O. citriodorum had a similar amount of oil synthesized in the leaves, however its fresh flowers revealed an oil content of 0.25%.

After water steam distillation, the main components of the essential oils were identified by different methods (GC, HPLC, GC-MS and co-chromatographies). The oil of leaves from O. sanctum contained predominantly, 1,8-cineol (11%), eugenol (10%), methylchavicol (7%), cadinene (7%) and humulene (3%), whereas the oil from blossoms was composed mainly of limonen, eugenol,  $\beta$ -caryophyllene, humulene and cadinene (in this sequence). The oil from leaves of O. citriodorum was characterized by limonene, linalool, citral and eugenol, and oil from blossoms by nerol, linalool, citral, geraniol and  $\beta$ -caryophyllene.

The process of drying resulted in a loss of essential oil by more than 50%.

Ocimum essential oil revealed a potent antimicrobial action. It could be demonstrated that terpenoids mainly interfered with respiratory and photosynthetic electron transport, concomitantly occurring proton translocations and coupled phosphorylation steps. Specific sites of action could be located by comparative use of well defined inhibitor and/or uncoupler substances (as ref. see 1).

- (1) K. Knobloch, H. Weigand, N. Weis, H.-M. Schwarm and H. Vogenschow, Progr. Essential Oil Research, Proc. 16th Internatl. Sympos. Essential Oils, Ed. E.-J. Brunke, Walter de Gruyter & Co, Berlin, New York (in press).

23 | CONSIDERATION OF MORPHOLOGICAL PHENOMENA AT  
 IN VITRO CLONAL MULTIPLICATION OF DIGITALIS SPECIES  
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It is a fact that *Digitalis* species show great variability in morphology, yielding capacity and chemical composition during their cultivation in the field. In order to eliminate this kind of a variability, a method called "Shoot Clonal Multiplication" can be used. The method is based on repeated induction of shoots from axillary buds in sterile culture/on Linsmaier and Skoog's medium supplemented with growth hormones/ and reconstitution of functional plants by rooting the shoots. After the adaptation of these plants in the green-house they can be transferred to the open. According to references by this method it is feasible to maintain large uniform population of heterozygous plants, favourably with high glycoside content /1,2,3/.

However, as a result of our experiments, we realized that if a growth hormone is applied for an extended period of time during subculturing of plants, morphological variability occurs even at the state of shoot cultures. These morphological variants often seem to be stable. Our protein investigation by polyacrylamide gel electrophoresis performed from the shoots used for propagation and leaves of plants with various morphological phenomena show qualitative and quantitative differences. This seems to indicate different gene expressions although we are always operating with standardized external circumstances. As morphological variability still occurs, this could be a result of a hormone-effect, which influences the plantlets in various ways. All these might result in an irreversible change of morphological characters.

1. Erdei, I., et al., 1981. Plant Cell Reports 1:34-35
2. Dobos, É., et al., 1982. Herba Hung. 21:49-57
3. Luckner, M., et al., 1984. Int. Vortragstagung, Artern, Vortragstexte Teil I:113-127



24 | ZUR UNTERSUCHUNG DER PHARMAZEUTISCH GENUTZTEN  
INHALTSSTOFFE VON VALERIANA OFFICINALIS L.

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Rohstoffe für pflanzliche Arzneimittel nur aus der Quelle der Wildsammlung zu gewinnen, wäre ökologisch unverantwortlich. Es wird daher ein landwirtschaftlicher Anbau angestrebt.

Eine der von uns untersuchten Pflanzen ist Valeriana officinalis L. Bekanntlich sollen hieraus hergestellte Präparate dazu beitragen, häufig auftretende Folgen von Streß, wie z.B. nervöse Unruhe und nervös bedingte Schlaflosigkeit zu mildern.

Als Fortsetzung früherer Untersuchungen wurde in den Jahren 1983/84 ein Versuch zur Ontogenese von Valeriana officinalis L. durchgeführt. Die unterirdischen Organe von jeweils 20 Einzelpflanzen wurden zu 15 Zeitpunkten während der Entwicklung der Pflanze geerntet. Die Analyse des ätherischen Öles geschah vorläufig durch Wasserdampfdestillation. Der Gesamtvalepotriatgehalt wurde nach Dichlormethanextraktion mittels HPLC aufgetrennt.

Der Verlauf der Kurve während der 2jährigen Entwicklung der Pflanze zeigt, daß die höchsten Erträge relativ spät im Jahr geerntet werden (Oktober) und daß hohe Gehalte an Valepotriaten und ätherischem Öl nicht nur im Herbst, sondern auch während eines genau definierten Zeitpunktes im Frühjahr auftreten. Das bedeutet für die landwirtschaftliche Praxis eine Verteilungsmöglichkeit für Ernte und Aufarbeitung der Droge.

Es wird außerdem eine Stadienskala für die Entwicklung von Valeriana officinalis L. vorgeschlagen, die vor allem dazu dienen soll, ohne aufwendige Untersuchungen der unterirdischen Masse, schon aus dem Erscheinungsbild der Pflanze den optimalen Erntezeitpunkt erkennen zu können.

26 | RADIOIMMUNOASSAY FOR THE DETERMINATION OF INDOLE  
ALKALOIDS IN CATHARANTHUS ROSEUS

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The purpose of this investigation was to find out whether vincristine content can be quantified from Catharanthus roseus using radioimmunoassay (RIA) (1,2). This alkaloid has been employed for many years as a chemotherapeutic agent in the treatment of neoplastic diseases.

Vincristine-protein conjugates were prepared by initial diazo coupling of p-aminophenylalanine or p-aminobenzoic acid to the 12'-carbon of the catharanthine moiety, followed by carbodiimide conjugation to protein amino groups. The purified and lyophilized immunogen was given in subcutaneous injections to four rabbits at four-week intervals.

The RIA-procedure was based on polyethylene glycol precipitation of the antigen-antibody complex, and liquid scintillation counting of the tritiated free antigen.

The sensitivity of the method was approximately 50 pg in 100  $\mu$ l samples. The crossreaction percentages of vinblastine and vindoline were 0,03 % and 0,09 %, respectively, and no interference was observed for 11 other compounds tested. Recovery of vincristine added to an extract pool from C. roseus was  $101,04 \pm 4,25$  % (n=12). Within-assay and between-assay variations were 4,7 % (n=30) and 7,1 % (n=15), respectively. The regression equation was  $y = 0,93 + 1,01x$ , and the coefficient of correlation was  $r = 0,9996$  (n=12).

The RIA-method described here has a good combination of high titer, sensitivity and specificity, and it appears to be very suitable for quantification of indole alkaloids from plant material.

- (1) T. Lehtola, A. Huhtikangas, R. Hiltunen and M. von Schantz, *Planta Medica*, 39 (1980) 273
- (2) T. Lehtola, A. Huhtikangas and R. Virtanen, *Planta Medica*, 45 (1982) 237

27 | DETERMINATION OF DIGITALIS LANATA CARDENOLIDES  
IN PLANT MATERIAL BY HPLC

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An HPLC method can be used to screen for the production of the A and C series of digitalis glycosides in Digitalis lanata plant material. For this purpose several different reversed phase combinations were tested (1). The most useful combination proved to be an analytical C<sub>18</sub> column (250 x 4.6 mm I.D., 5 µm) together with a C<sub>2</sub> precolumn (40 x 4.6 mm I.D., 10 µm).

The qualitative and quantitative effectiveness of the method were tested with standards. The reproducibility of the peak heights and retention times was determined for 17 cardenolides with 6 parallel runs. The minimum detection limits for A and C glycosides were from 5 to 15 ng. The linearity of the detection as function of peak height was also determined. The applicability of the method was tested with plant materials of different origin.

The results show that the method is useful for screening Digitalis lanata plant material on the basis of the production spectrum of cardenolides.

- (1) T. Laakso, V. Kauppinen & R. Joensuu,  
Farm. Tijdschr. Belg., 61 (1984) 360.



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In the last few years several 5,6-dihydroxy-7,8-dimethoxyflavones and 5,8-dihydroxy-6,7-dimethoxyflavones have been isolated and identified (1-3). The characterization of these compounds was not easy by means of classical techniques. The obtaining of Wessely-Moser isomers by acidic treatment have proved to be quite useful for the identification of these rare flavonoids (2). In this work we show UV, EIMS and chromatographic evidences to differentiate clearly these isomeric compounds. The study of the UV spectra in MeOH of the isomeric couples showed the following differences (4): A) The 5,8-dihydroxyflavonoids exhibited a BIII (295-325 nm) (maximun, shoulder, inflection) which is absent in 5,6-dihydroxyflavonoids spectra. B) Flavonoids with hydroxyl at C-8, generally showed shorter wavelenghts values for BI and BII maxima than their isomeric compounds with hydroxyl groups at C-6.

The shape of the  $AlCl_3 + HCl$  spectra of 6-hydroxy-8-methoxycompounds are characterized by two principal absorption bands, the BIb and BIIa. On the other hand, the spectra of 6-methoxy-8-hydroxycompounds exhibited three or more bands between the BIIa and BIb.

It is noteworthy that the 5,8-dihydroxyflavones decompose much more quickly than the 5,6-dihydroxyflavones. This decomposition is increased in alkaline media, and it is clearly evidenced in the NaOMe-UV spectra by the decrease in absorbance.

The EIMS evidenced that the presence or absence of the  $M-H_2O^+$  peak is very useful for distinguishing between these two isomers, being this ion absent in the spectra of 6-hydroxy-8-methoxyflavones, and present (ca 7% rel. abund.) in 6-methoxy-8-hydroxyflavones.

In addition, 6-hydroxycompounds showed lower  $R_f$  values in cellulose TLC with 30% or 60% HOAc, than the 8-hydroxy- isomers, and 6-hydroxyflavones eluted with shorter retention times in HPLC with reversed phase columns, than the 8-hydroxy- isomers.

- (1) C.O. Van den Broucke, R.A. Dommissse, E. L. Esmans and J.A. Lemli, *Phytochemistry*, 21 (1982) 2581.
- (2) F.A.T. Barberán, F. Tomás and F.Ferreres, *Phytochemistry*, 23 (1984) 2112.
- (3) F. Ferreres, F.A.T. Barberán and F. Tomás, *Phytochemistry*, 24 (1985) 1869.
- (4) F.A.T. Barberán, F. Ferreres and F. Tomás, *Tetrahedron*, in press.

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Die Ueberdruckschichtchromatographie (OPLC), entwickelt von Tyihak et al. (1), bei der der Dampfraum durch vollständige Bedeckung der Schicht mit einer unter Druck stehenden elastischen Membran praktisch eliminiert ist, wurde bis jetzt hauptsächlich für analytische Trennungen angewendet.

In der letzten Zeit wurde die OPLC in geringem Mass für präparative off-line Trennung verwendet und kürzlich für präparative on-line Trennungen (2) entwickelt. Unter Verwendung eines geeigneten Verteilungs- und Auslaufsystems wird die mobile Phase durch die an allen Rändern imprägnierte Adsorbenschicht geleitet. Die getrennten Substanzen können mit Hilfe eines Durchflussdetektors mit einem Schreiber registriert und mit einem Fraktionensammler aufgefangen werden.

Die umgebaute Chrompres-10 Kammer erlaubt die Verwendung von 20 x 20 cm und 20 x 40 cm Fertigplatten von 0,5 bis 2 mm Schichtdicke. Man kann zwischen 50-500 mg Probenmenge auftragen. Die Methode wird mit einigen Beispielen für schnelle Isolierungen und komplex zusammengesetzte Pflanzenextrakte demonstriert.

\* Zur Zeit am Pharm. Inst. der ETH Zürich, von der Medizinischen Universität Semmelweis, Institut für Pharmakognosie, H-1085 Budapest

#### Literatur

- (1) E. Tyihak, E. Mincsovcics and H. Kalasz: J. Chromatogr. 174, 75 (1979).
- (2) Sz. Nyiredy, C.A.J. Erdelmeier und O. Sticher: Instrumental Preparative Chromatography (Ed. R.E. Kaiser et al.), im Druck.

30 | ROTACHROM: Ein neues Instrument für on-line  
Isolierungen (SCLC, CPCC)

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ROTACHROM ist ein neues zentrifugal beschleunigtes Modularsystem zur Planar- Chromatographie, das drei verschiedene Trennmöglichkeiten bietet (1,2,3). Das Instrument beinhaltet eine Grundausrüstung, die für analytische und präparative Zentrifugal-Schicht-Chromatographie (CLC), für analytische und preparative Sequenzzentrifugal-Schicht-Chromatographie (SCLC) und präparative Zentrifugal-Planarsäulen-Chromatographie (CPCC) verwendet werden kann. Bei allen drei Methoden wird im zirkularen Modus getrennt, die Zentrifugalkraft kann durch den Motor reguliert werden.

Bei der SCLC können zirkularer und antizirkularer Modus so oft wie nötig kombiniert werden, damit kann die Trennstrecke theoretisch unendlich verlängert werden (4). Durch die zeitlich und örtlich variable Lösungsmittelzufuhr kann man komplexe Trennprobleme auf einer Platte lösen (5).

Die CPCC ist eine geschlossene zentrifugal-beschleunigte Planartechnik, die durch eine spezielle geometrische Form der Trennkammer ein konstantes Volumen des Adsorbens entlang des Radius sichert. Ohne Zusatz von Bindemittel kann mit jedem beliebigen Trennmateriale (z.B. RP-18 etc.) gearbeitet werden.

Die Probenmenge kann bei allen drei Methoden zwischen 50-500 mg betragen.

#### Literatur

- (1) Angemeldet für Internat. Patent, Reg. Nr. 1717/84 (1984).
- (2) Angemeldet für CH-Patent, Reg. Nr. 1588/85 (1985).
- (3) Angemeldet für CH-Patent, Reg. Nr. 1589/85 (1985).
- (4) Sz. Nyiredy, C.A.J. Erdelmeier and O. Sticher: HRC & CC 8  
(2/1985) 73.
- (5) C.A.J. Erdelmeier, Sz. Nyiredy and O. Sticher: HRC & CC 8  
(3/1985) 132.



## Ein substanzschonendes Isolierungskonzept für polare Naturstoffe

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Der Isolierung von empfindlichen Pflanzeninhaltsstoffen im genuinen Zustand sind bei Anwendung herkömmlicher chromatographischer Trennmethode, die vornehmlich auf der wiederholten Anwendung von offenen Säulen mit Kieselgel als Adsorbens beruhen, Grenzen gesetzt. Die irreversible Adsorption polarer Substanzen (Glykoside) sowie die Möglichkeit unkontrollierbarer katalytischer Veränderungen sensibler Stoffe stellen die klassische Methodik in Frage. Da Pflanzenextrakte in der überwiegenden Zahl der Fälle hochkomplexe Substanzgemische darstellen, ist ein Mehrschrittverfahren zur Vor- und Endreinigung der gewünschten Komponenten unumgänglich.

Im Rahmen der Untersuchung der Inhaltsstoffe des methanolischen Extraktes der Wurzeln von *Sambucus ebulus* L. (*Caprifoliaceae*) [1] wurde ein Isolierungskonzept entworfen, welches sich vom herkömmlichen Vorgehen durch die ausschliessliche Anwendung substanzschonender chromatographischer Trennverfahren und völligen Verzicht auf aggressive Adsorbentien wie Aluminiumoxid, Bleiacetat oder Kohle usw. zur Vorreinigung der Extrakte unterscheidet. Sein zentraler Gedanke ist die Kombination der an sich bekannten Trenntechniken

- Flüssig/Flüssig-Verteilung
- Gel filtration
- Umkehrphasenchromatographie

in der erwähnten Reihenfolge. Die präparative Gradientenniederdruckchromatographie an Reversed-Phase-Material als stationäre Phase wurde dabei erstmals konsequent eingesetzt. Zur Erzeugung der Elutionsgradienten wurde das einfache Prinzip der kommunizierenden Gefässe eingesetzt.

Die optimierte Technik erlaubte die ökonomische Isolierung von 14 Inhaltsstoffen aus dem methanolischen Extrakt der Wurzeln von *Sambucus ebulus*, wobei 9 der isolierten Substanzen bisher nicht beschriebene Pflanzeninhaltsstoffe darstellen. Dabei handelt es sich teilweise um äusserst empfindliche Iridoid- bzw. Secoiridoidmonoglykoside und ein Iridoiddiglykosid vom Valerianatyp sowie neuartige, mit Arbutin verwandte monomere Phenol- und ein dimeres Phenylpropanoidglykosid. Ueber die Konstitutionsaufklärung zweier der isolierten Substanzen wird an diesem Kongress in Form von Postern berichtet.

Die Einsatzmöglichkeiten der Methode erstrecken sich auf phytochemische/chemotaxonomische Untersuchungen im Grundlagenforschungsbereich wie auch, bei entsprechendem Upscaling, auf semiindustrielle Isolierungsarbeiten.

Literatur: [1] G.A.Gross, Diss ETH Zürich Nr.7800, 1985.

Einsatz der RP-HPLC gekoppelt mit EC- und/oder  
UV-Detektor zur quantitativen Analyse der l-Ascorbinsäure  
in Pflanzen am Beispiel von *Cynobati fructus*

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Verschiedene Techniken zur quantitativen Bestimmung der l-Ascorbinsäure in Pflanzen wurden entwickelt, sind jedoch in mehreren Punkten unbefriedigend. Klassische Methoden wie Redox titrationen mit Jod oder anderen Reagenzien sind wenig spezifisch und geben in gefärbten Lösungen Probleme bei der Endpunktbestimmung. Spektrophotometrische, mikrofluorimetrische und polarographische Methoden werden u.a. ebenfalls eingesetzt, sind jedoch teilweise wenig empfindlich oder aufgrund der komplexen Pflanzenmatrix störanfällig, teilweise in ihrer Probenaufbereitung kompliziert und zeitaufwendig. Ziel der Arbeit war daher die Entwicklung einer möglichst einfachen, v.a. jedoch selektiven Analysenmethode.

Die RP-HPLC bietet aufgrund einfacher Handhabung, hoher Trennleistung und für die Quantifizierung geeigneter Detektionssysteme klare Vorteile. Durch stark saure wässrige mobile Phasen (0.5 % m-Phosphorsäure, pH 2) wird das Retentionsverhalten der polaren Ascorbinsäure günstig beeinflusst ("Hydrophobic Chromatography"), so dass auf ionenpaarbildende Reagenzien verzichtet werden konnte. Der Einsatz eines elektrochemischen Detektors (1), der sich - wenn auch kürzlich noch umstritten - zusehends in der HPLC etabliert, bringt für spezielle Probleme der Pflanzenanalytik - wie z.B. die Ascorbinsäurebestimmung - Vorteile bezüglich Selektivität und Empfindlichkeit. Verschiedene Methoden (EC- und UV-Detektion, externer und interner Standard) wurden miteinander verglichen.

Die Probenaufbereitung - aufgrund der Instabilität wässriger Ascorbinsäurelösung ein wichtiger Punkt - konnte unter Verwendung von Bond Elut C18 Einweg-Extraktionssäulen optimiert werden, so dass dank minimaler Belastung der analytischen Säule Reihenanalysen innerhalb kürzester Zeit ohne Zwischenspülung möglich sind.

Verschiedene Drogen- und Teemuster des Handels sowie Proben von Frischpflanzen wurden analysiert, die Resultate (Ascorbinsäuregehalt 0.1 - 1.1 %) statistisch ausgewertet.

Literatur:

- 1) Stulik, K. und Pacakova V., CRC Crit.Rev.Anal.Chem. 14, 297 (1984)

33 | CURCUMIN- Chemistry, stability and analysis  
of a naturally occurring drug molecule.

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Curcumin is the main colouring principle found in the rhizomes of the plant Curcuma longa L. (Zingiberaceae). The rhizomes usually contain 2-5% of curcumin and two related demethoxycompounds present in small amounts; demethoxy- and bisdemethoxycurcumin. Curcumin and the curcuminoids belong to the group of diarylheptanoids. Even though curcumin has a long tradition as a colouring agent in food processing, cosmetics and textiles and is reported to be pharmacological active, little is known about the stability and chemistry of the molecule. It was therefore of interest to study the chemical and physical properties of the curcumin molecule, if possible stabilize it as a dye and further make use of it as a future drug/drug model. Methods: X-ray diffraction for determination of absolute structure. HPLC for analytical purposes, fluorimetry and UV/vis spectrometry.

Results: Curcumin is isolated and synthesized; absolute structure investigated, analytical method for quant./qual. determination developed and pH-dependant/photochemical degradation investigated.

Ref.: H.Hjorth Tønnesen, J.Karlsen

- Acta Chem. Scand., B 36 (1982) 475
- J. Chromatog., 259 (1983) 367
- Z.Lebensm.-Unters.-Forsch., 177 (1983) 348
- Ibid. 180 (1985) 132
- Ibid. 180 (1985) 402



34 |  $^1\text{H}$ - AND  $^{13}\text{C}$ -NUCLEAR MAGNETIC RESONANCE  
SPECTROSCOPIC ANALYSIS OF THE PYRROLIZIDINE ALKALOID  
CONTENT OF SENECIO VULGARIS, SENECIO VERNALIS AND  
SENECIO JACOBAEA

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An analytical method was developed in order to obtain qualitative and quantitative information on the pyrrolizidine alkaloidal constituents of Senecio vulgaris, S.vernalis and S.jacobaea, using  $^1\text{H}$ - and  $^{13}\text{C}$ -NMR spectroscopy.  $^1\text{H}$ -NMR appeared to be most useful for the determination of the total alkaloid level by comparison with an internal standard (1), and  $^{13}\text{C}$ -NMR was an excellent method to determine the relative amount of the different pyrrolizidines in the mixture, even if their chemical structures were very similar. For the optimization of the quantitative  $^{13}\text{C}$ -NMR measurements, it was necessary to measure the most important spin-lattice relaxation times. In spite of the instrumental and experimental difficulties which had to be overcome the precision of the results produced by  $^{13}\text{C}$ -NMR is good (0,5 - 1,5 %) (2). Some samples were analysed by HPLC (3) and GC/MS as well, but the combination of quantitative  $^1\text{H}$ - and  $^{13}\text{C}$ -NMR has one important advantage viz. Structural information and quantitative results are obtained simultaneously. In general this technique can be used to analyse all kinds of complex mixtures of related compounds, in a nondestructive way.

1. Pieters L.A.C. and Vlietinck A.J.: Fresenius Z.Anal. Chem. 321, 355-358 (1985)
2. Pieters L.A.C. and Vlietinck A.J.: submitted for publication in 'Magnetic Resonance in Chemistry'(1985)
3. Pieters L.A.C. and Vlietinck A.J.: submitted for publication in 'Journal of Liquid Chromatography'(1985)

\* L.A.C.Pieters is a research assistant of the Belgian Fund for Scientific Research (NFWO).

35 | THE RELIABILITY OF SIMILARITY INDICES FOR COMPARING  
SPECTRAL DATA IN THE GC-MS ANALYSIS OF TERPENES

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The identification of compounds based on computer matching against a library data is an important technique in volatile oil analysis by GC-MS (1-2). For the creation of an own reference data the analytical conditions in terpene analysis were investigated and comparisons were made using the similarity index.

A quadrupole MS (HP 5970A) coupled to an HP 5890 GC was used under the control of an HP 9825B Desktop Computer with an HP 9134A Disc Memory for data storage. The analyses were performed on an OV-351 vitreous silica column under following MS conditions: scan rate 690 amu/sec, ion source 70 eV, electron multiplier energy 1600 V, vacuum  $1 \times 10^{-5}$  torr. The 10 most significant fragments for each compound were stored in the library file.

The mean reproducibility of the 10 most significant peaks of a monoterpene ( $\alpha$ -thujene) in GC-MS was 3.7% and 3.2% (C.V., N=6) at the total abundance (TA) range of 1500 and 3000 respectively decreasing drastically at the range less than 500 (5-15%, C.V.). The precision for fenchylalcohol and farnesol was 5.3% and 9.8% (C.V.) at the respective TA levels of 5000 and 1200. The similarity index for these compounds was poor ( $S_I=0.960-0.980$ ) since the 10 fragments were not always the same.

Comparing the similarity between two co-eluting compounds with similar spectra profiles, such as  $\alpha$ -pinene and  $\alpha$ -thujene, indicated that a high index for  $\alpha$ -pinene (0.999) was unaffected when the amount of  $\alpha$ -thujene was increased to 50% of the  $\alpha$ -pinene content. The spectrum of geranylacetate ( $S_I=1.000$ ) was disturbed when 15% of citronellol was added, an index of 0.998 being obtained.

At low TA range for carvacrol (200-400), thymol was proposed as the first match with a high index (0.991-0.992). Misclassifications were also found between l- and isoborneol, and geranyl- and nerylacetate. The precision at low abundance levels was found to be insufficient for reliable application of the similarity index. If the fragmentation patterns are similar, then a substantial amount of co-eluting compound can be present without any drastic decrease in the index value. When replicate analyses of alcohols were carried out, the 10 fragments were not always the same as stored in the data library. Incorrect matches were also obtained between isomers, although if the exact retention is known then most of the errors can be eliminated.

(1) Knock et al., Anal.Chem. 42 (13) (1970) 1516.

(2) Adams et al., J.Chromatogr.Sci. 17 (1979) 75.

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Das Iridoidglukosid Aucubin aus den Spitzwegerichblättern (*Folia Plantaginis lanceolata*) ist, vergleichbar den als Iridoidestern vorliegenden Valepotriaten aus der Baldrianwurzel, ein sehr labiler Pflanzeninhaltsstoff. Aucubin eignet sich somit als Indikator für die Beurteilung der Drogengewinnung und -aufarbeitung. Erstes Ziel der vorliegenden Arbeit war deshalb die Ermittlung des Aucubingehaltes in Blatt- und Krautdroge von *Pl. lanceolata* sowie in dem verschiedentlich als Ersatzdroge angegebenen Breitwegerichkraut (*Pl. major*). Darüberhinaus wurde untersucht, wieviel Aucubin in eine wässrige Zubereitung übergeht bzw. in entsprechenden Fertigarzneimitteln enthalten ist. Die mittels HPLC durchgeführte quantitative Bestimmung ergab, daß die Blattdroge von *Pl. lanc.* einen deutlich höheren Aucubingehalt aufwies als die Krautdroge, der Gehalt der Herbadroge von *Pl. major* lag jedoch noch weit darunter. In wässrigen Zubereitungen wurden ca. 70 % Aucubin wiedergefunden, die damit einen wesentlich höheren Aucubingehalt pro Einzeldosis haben als vergleichbare Fertigarzneimittel.



# ANALYSIS OF PYRROLIZIDINE ALKALOIDS IN EUPATORIUM CANNABINUM L. BY MEANS OF POSITIVE AND NEGATIVE ION CHEMICAL IONIZATION GC-MS

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The combination of positive and negative ion chemical ionization (CI) in gas chromatography-mass spectrometry of trimethylsilyl (TMS) derivatives of pyrrolizidine alkaloids (PAs) offers a rapid, tentative structure elucidation of suchlike compounds in plant material [1]. Positive ion CI with  $\text{NH}_3$  as the reactant gas gives abundant  $\text{MH}^+$  ions. Elimination of the necic acid produces a positive ion, corresponding with the necine base, which may carry a substituent at  $\text{C}_7$ . Negative ion CI with  $\text{OH}^-$  as the reactant ion cleaves the ester bonds which may be present at  $\text{C}_7$  and  $\text{C}_9$  of the necine base or at the  $\beta$ -C atom of the necic acid.

at the  $\beta$ -C atom of the necic acid.

Compounds identified in aerial parts of Eupatorium cannabinum L. are four echinatine isomers (o.a. lycopsamine and intermedine) and some of their  $\beta$ -acetyl,  $\beta$ -angelyl/tiglyl and  $\beta$ -(iso)valeryl esters. PAs without a substituent at  $\text{C}_7$  were tentatively identified as supinine and amabiline.

In addition to a number of these alkaloids, some  $\beta$ -(iso)butyl,  $\beta$ -angelyl/tiglyl and  $\beta$ -(iso)valeryl esters of supinine or amabiline were detected in subterranean parts of the plant. PAs with a saturated necine base like three trachelanthamine isomers and some  $\beta$ -angelyl/tiglyl esters could be detected in the root material only. A  $\text{C}_9$ -viridifloryl/trachelanthyl ester of a saturated amino alcohol like turneforcidine and one of its  $\beta$ -angelyl/tiglyl esters had also been found.

The latter 2 compounds, the  $\beta$ -(iso)butyl, the  $\beta$ -(iso)valeryl and the  $\beta$ -angelyl/tiglyl esters of supinine or amabiline and the  $\beta$ -(iso)valeryl ester of an echinatine isomer have not been described in nature before.

- [1] H.J. Huizing, F. de Boer, H. Hendriks, W. Balraadsingh and A.P. Bruins (in preparation).

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The isolation and identification of chemical compounds of the rhizoma of Pakisnaga ( Polypodium Feei Mett., Polypodiaceae) were carried out. The preliminary chemical study showed the presence of tanin, flavonoid and triterpenoid.

A pentacyclic triterpene which was supposed to be arbor-7-ene was isolated from the petroleum ether extract and identified by UV and IR spectrophotometry, mass spectrometry, RMN spectrometry and CHNS analysis.

From the n-butanol extract a flavonoid could be isolated which was assumed to be a glycoside of kaemferol derivative with two sugars.

39 | DENSITOMETRISCHE GEHALTSBESTIMMUNG VON ALOE-  
EMODIN-GLYKOSIDEN IN SENNESFRÜCHTEN UND  
-BLÄTTERN UND DEREN WÄSSRIGEN ZUBEREITUNGEN

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Sennesfrüchte (Sennae fructus angustifoliae und S. fructus acutifoliae Ph. Eur. I) gelten im Vergleich zu Sennesblättern (S. folium Ph. Eur. I) trotz eines höheren Anthraglykosidgehalts als milderer und mit weniger Nebenwirkungen behaftetes Laxans (1,2,3). Dieses wird u.a. mit einem geringeren Aloe-emodin-glukosid-Gehalt erklärt (4). Da in der Literatur die Angaben für die Aloe-emodin-glukosid-Gehalte variieren, war das Ziel der vorliegenden Untersuchung, die Schwankungsbreite der Aloe-emodin-glykosid-Gehalte sowohl in Blättern und Früchten der beiden unterschiedlichen Sennesherkünfte als auch in deren wässrigen Zubereitungen zu bestimmen. Dazu wurde eine densitometrische Gehaltsbestimmung für Aloe-emodin- und Rhein-glykoside erarbeitet.

- (1) P.-J. Schorn, W. Schmid in Ph. Eur. I Kommentar 2. Auflage, Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart 1978, S. 1153
- (2) R. Hänsel, H. Haas, Therapie mit Phytopharmaka, Springer Verlag, Berlin, Heidelberg, New York, Tokyo 1983, S. 163
- (3) M. Luckner, O. Bessler und R. Luckner, Pharmazie 22, 379 (1967)
- (4) J.W. Fairbairn, Planta med. 7, 406 (1959)



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Within the scope of an investigation of sesquiterpene lactones from Eupatorium species, high-performance liquid chromatography on octadecyl-silica (C-18) stationary phase offers an efficient and rapid method for analysis of sesquiterpene lactones.

A mixture of known sesquiterpene lactones, isolated from Eupatorium cannabinum L. (1), was analysed with acetonitril/water gradient as described by Strack and co-workers (2).

Using a Diode-Array-Detector and gradient elution it is possible to detect three types of components; sesquiterpene lactones, flavonoids and another type of compounds, unknown so far. These unknown compounds may be chromenes or benzofurans, in view of the results of Lins and co-workers (3). The presence of sesquiterpene lactones and flavonoids was demonstrated by co-chromatography of selected reference compounds using both retention-times and UV-spectral data.

The identity of the known reference compounds was confirmed by IR, H-NMR, <sup>13</sup>C-NMR and Mass-Spectrometry.

- (1) Bos, R., Hendriks, H., Bruins, A.P., Schripsema, J., Kloosterman, J. and Sipma, G. *Farmaceutisch Tijdschrift voor België*, 61 398 (1984).
- (2) Strack, D., Proksch, P. and Gülz, P.G. *Z. Naturforsch.* 35c G15 (1980).
- (3) Lins, R., Palmer, J., Proksch, P. Poster presented on the 33<sup>rd</sup> Annual Congress on Medical Plant Research, Regensburg 1985.

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Nach dem Stand der bisherigen Erkenntnisse beruht die sedative Wirkung der Baldrian-Drogen und ihrer Zubereitung nicht auf eine Wirkstoffklasse alleine, sondern möglicherweise auf dem Zusammenspiel mehrerer Stoffklassen.

Wesentlich an der Wirkung beteiligt sind die Valepotriate (1), ihre Abbauprodukte (2) und die Bestandteile des etherischen Öls (3). Im Überblick aller bisher bekannten Analysenverfahren kann festgestellt werden, daß das HPLC-Verfahren für die nicht-flüchtigen Anteile und die Kapillargaschromatographie für die flüchtigen Anteile von Valeriana-Drogen am Besten geeignet sind.

Es sollten alle wichtigen Wirkstoffe und deren Abbauprodukte, sowie die Leitsubstanzen nebeneinander in einem HPLC-Analysengang mit Dioden-Array-Detektion untersucht werden. Ebenso sollte mit den jeweiligen etherischen Ölen und der gaschromatographischen Analyse verfahren werden.

Mit der HPLC-DAD-Methode wurden mehrere Proben Valeriana off. untersucht. In Bestätigung früherer Untersuchungen (4) wurden in allen Val. off. Proben Valerensäure-derivate gefunden.

Wesentlicher Vorteil der HPLC-Methode ist die direkte Bestimmung der Isomerenverteilung zwischen Didrovaltrat und Homodidrovaltrat, sowie zwischen Valtrat, Isovaltrat und Homovaltrat.

Ebenso wie der chromatographische Fingerprint kann auch das Valtrat Isomerenverhältnis nur sehr bedingt als Identifizierungsmerkmal chemischer Rassen herangezogen werden, da es bei den hier beschriebenen Untersuchungen sehr unterschiedlich war.

Anhand der GC-Analysen konnten die Valerenan-Sesquiterpenoide (5) nachgewiesen werden, die zum Teil als Leitsubstanz dienen können, weil sie bis jetzt nur in Val. off. nachgewiesen worden sind. Außerdem kann man anhand der qualitativen Zusammensetzung verschiedene Öl-Typen unterscheiden: 1<sup>e</sup> ein Valerenal-Typ; 2<sup>e</sup> ein Valeronon-Typ; ein Sesquiterpenalkohol-Typ und ein Ketovaleronon-Typ.

1. Eickstedt, K.W. von, Rahmen, S., *Arzneim. Forsch.* 9, 316 (1969)2. Wagner, H., Jurcic, K., Schaette, R., *Planta Med.* 39, 358 (1980)3. Hendriks, H., Bos, R., *Dragoco report*, 31, 3 (1984)4. Hänsel, R., Schulz, J., *Dt. Apoth. Ztg.* 122, 215 (1982)5. Bos, R., Hendriks, H., Kloosterman, J., Sipma, G., *Phytochem.* accepted for publication

GLYKOSIDE MIT WASSERDAMPFFLÜCHTIGEN GENINEN IM BLATT  
VON MELISSA OFFICINALIS L.

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Im Rahmen einer umfangreicheren phytochemischen Untersuchung der Inhaltsstoffe von Melissenblatt haben wir die Anwesenheit von Iridoiden und von Glykosiden mit flüchtigem Genin überprüft. Die Suche nach den Iridoiden verlief negativ (1).

Der Methanolextrakt der Droge wurde gereinigt, indem er mit Petrolether ausgezogen wurde. Dann wurde er folgendermassen chromatographisch weiterbehandelt : zuerst über Aluminiumoxid (Wasser), anschliessend über Aktivkohle (Wasser-Ethanol) und am Schluss über Silicagel (Ethylazetat und Methanol). Das so gereinigte Extrakt wurde mit  $\beta$ -Glucosidase (aus Mandeln) in Phosphat-Puffer bei pH 5 inkubiert. Als einziger Zucker wird bei der enzymatischen Hydrolyse die Glucose freigesetzt. Sie konnte durch DC und durch GC nach Silylierung identifiziert werden. Bei dieser Spaltung entstehen sieben Aglykone : Benzylalkohol,  $\beta$ -Phenylethylalkohol, Eugenol, Nerol, Geraniol, Nerylsäure und Geranylsäure. Sie wurden kapillarchromatographisch nach ihren Retentionszeiten und mittels ihren Massenspektren identifiziert.

(1) O. Sticher und M. Junod-Busch, Pharm. Acta Helv., 50 (1975) 127.



WASSERDAMPFFLÜCHTIGE BESTANDTEILE IN DEN BLÄTTERN VON  
LYCIUM HALIMIFOLIUM MILLER

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Lycium halimifolium MILLER (Solanaceae-Solaneae-Lyciinae WETTSTEIN)

ist eine in Europa weitverbreite Pflanze, die für ihre toxischen Eigenschaften bekannt ist (1).

Bei der Suche nach den toxischen Prinzipien haben wir, unter anderem, die flüchtigen Inhaltsstoffe der Blätter untersucht. Aus ungefähr 500 g getrocknetem Blattmaterial (Herkunft Wallis, Schweiz) konnten wir etwa 0,01% etherisches Öl destillieren (Apparatur der Ph.Eur.). Aus der qualitativen Analyse mittels GC-MS (Finnigan Databank, FIRMENICH AG, Genf) konnten wir ersehen, dass sich das etherische Öl aus mehreren hundert Bestandteilen zusammensetzt. Wir haben 133 davon identifiziert. Dabei handelt es sich hauptsächlich um Kohlenwasserstoffe, Alkohole und Oxoderivate, unter denen sich eine grosse Anzahl Verbindungen vom Jonon-Typ finden. Hauptbestandteil ist das Damascenon (1-[2,6,6-Trimethyl-1,3-cyclohexadien-1-yl]-2-buten-1-on), eine Verbindung, die von der Parfümerie her bekannt ist.

(1) D. Frohne und H.J. Pfänder, Giftpflanzen, Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart 1982, S. 209.

44 | SEDANOLIDE, A MAJOR COMPONENT IN  
CELERY OIL

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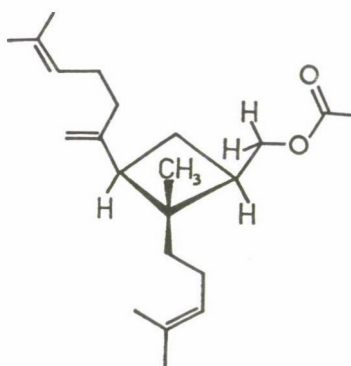
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The fruit oil composition of Apium graveolens L., cultivated at Mansoura, Egypt, was investigated. Identification of the components was mainly performed by means of GC/MS. About 20 components were identified. The oil composition, in general, is in accordance with that reported for oils collected from different parts of the world. However, only small amount of n-butyl phthalide (0.3%) is present while n-butanalidene phthalide and sedanenolide are absent. Surprisingly, a significant amount (14.5%) of sedanolide could be identified.

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The fruits of Ammi visnaga (L.) Lam. (Apiaceae) are of pharmaceutical importance because of their content of furanochromones and pyranocoumarine esters. In addition they contain 0.03% of an essential oil, which has recently been the subject of our chemical investigation (1). Besides some common monoterpenoids and aliphatic esters, a diterpenoid ( $C_{22}H_{36}O_2$ , m.w. 332) was isolated, which was proved to be an acetic ester of a diterpenoid alcohol. By means of two dimensional NMR spectroscopy, the structure of this diterpenoid could be established.



It can be considered as a cyclisation product of the monoterpenoids myrcene and geranyl acetate.

(1) Elisabeth Stahl (1983), GIT-Supplement 3/83, 69-72.



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*Salvia*-Arten wurden schon zur Zeit der Römer für arzneiliche Zwecke benutzt. Blätter, Blütenstände und die ätherischen Öle der *Salvia*-Arten finden Verwendung in der Pharmazie, Lebensmittel-, Gewürz- und Parfüm-Industrie (1, 2, 3, 4).

In Anatolien finden sich 86 wild wachsende *Salvia*-Arten. Die ätherischen Öle einiger Arten aus Anatolien sind in früheren Arbeiten beschrieben worden (6. 7. 8. 9). Unter diesen Arten ist *Salvia multicaulis* Vahl eine Art, die vorwiegend in Ostanatolien und den näheren süd- und zentralanatolischen Gebieten wächst (5). In dieser Arbeit wurden die Monoterpenoide des ätherischen Öles von *S. multicaulis* ermittelt.

Das Pflanzenmaterial für diese Arbeit wurde in der Umgebung von Antakya (Südtürkei) gesammelt. Das ätherische Öl wurde durch Destillation in einem Clevenger-Apparat isoliert. Das Öl wurde an einer Kieselgel-Säule fraktioniert. Und die Fraktionen wurden mittels Gaschromatographie untersucht (10).

Die Ausbeute an ätherischem Öl war 0,6 %. Die Hauptkomponente des Öls ist Campher.

Andere analytische Daten und die Gaschromatogramme werden praesentiert.

- (1) Perrot, E., Paris, R.: *Les Plantés Médicinales*, Presses Universitaires de France, Vendome, 1971.
- (2) Valnet, J.: *Aromathérapie*, Librairie Meloine S.A. Editeurs, Paris, 1972.
- (3) Duquenois, P.: *Quarterly J. Crude Drug Res.*, 12(1), 1841 (1972).
- (4) Bardeau, F.: *La Pharmacie du bon Dieu*, Editions Stock, Paris (1973).
- (5) Davis, P. H.: *Flora of Turkey and the East Aegean Islands Vol.7*, Edinburg University Press, Edinburg, 1982.
- (6) Tanker, M., Sarer, E., N.: *J. Fac. Pharm. Ankara*, 6, 198 (1976).
- (7) Sarer, E.: *J. Fac. Pharm. Ankara*, 10, 112 (1980).
- (8) Sarer, E.: *J. Fac. Pharm. Ankara*, 13, 146 (1983).
- (9) Sarer, E.: *J. Fac. Pharm. Ankara*, im Druck
- (10) Sarer, E.: *J.J.C., Svendsen, A.B.: Sci. Pharm.*, 51 58 (1983).

THE ESSENTIAL OILS OF THREE ORIGANUM SPECIES GROWN IN  
TURKEYE. Şarer<sup>1</sup>, J.J.C. Scheffer<sup>2</sup>, A. Looman<sup>2</sup> and A. Baerheim Svendsen<sup>2</sup><sup>1</sup>Faculty of Pharmacy, Ankara University, Tandogan-Ankara, Turkey<sup>2</sup>Division of Pharmacognosy, Center for Bio-Pharmaceutical Sciences,  
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Origanum species have been used in medicine and as spices ever since antiquity, mainly because of their content of essential oils. In previous studies we investigated the essential oil of O. majorana grown in southern Turkey (1) and we studied its antimicrobial activity.

For the present study we collected flowering parts of O. onites L., O. syriacum L. var. bevanii (Holmes) Ietswaart and O. vulgare L. ssp. hirtum (Link) Ietswaart. The plant material was identified by Ietswaart who made a taxonomic revision of the genus Origanum (2).

About 1 kg of dried material of each specimen was collected. The flowering parts were subjected to hydrodistillation for 3 h, which yielded 2-3% of essential oil. The oils were analyzed by GC using 60 m fused silica capillary columns.

The analysis of the three oil samples showed that they were characterized by a high content of carvacrol (61-72%). Other oxygen-containing components were only present in minor amounts, except linalool that amounted to 5% in O. onites oil. Two monoterpene hydrocarbons were found in relatively large amounts:  $\gamma$ -terpinene (3-8%) and p-cymene (6-13%).

Further analytical data and the results of an antimicrobial screening carried out by means of the agar overlay technique as described elsewhere (3) will be presented.

- (1) E. Şarer, J.J.C. Scheffer and A. Baerheim Svendsen, *Planta Med.*, 46 (1982) 236
- (2) J.H. Ietswaart, A taxonomic revision of the genus Origanum (Labiatae), Leiden University Press, The Hague/Boston/London, 1980
- (3) A.M. Janssen, J.J.C. Scheffer, A. Baerheim Svendsen and Y. Aynehchi, *Pharm. Weekbl. (Sci.)*, 6 (1984) 157

CHROMENE AND BENZOFURAN METABOLISM IN  
AGERATUM HOUSTONIANUM AND EUPATORIUM  
ADENOPHORUM (ASTERACEAE)

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Chromenes and benzofurans are characteristic and biologically active constituents of the Asteraceae (1). Ageratum houstonianum and Eupatorium adenophorum were chosen as model systems to gain first insights into the physiology and metabolism of this class of natural products. A. houstonianum accumulates the antijuvenile hormones precocens I and II largely in the mesophyll of the leaves. Minor amounts in comparison are found in stems and roots. Benzofurans are confined solely to the roots where they are stored in resin ducts. E. adenophorum elaborates several chromenes that are also stored largely within the mesophyll of the leaves. All compounds, especially those from E. adenophorum, are subject to a fast turnover during seed germination and early seedling development. The possible biogenetic conversion of the compounds as indicated by their different accumulation maxima during early ontogeny is discussed.

- (1) P. Proksch and E. Rodriguez, *Phytochemistry*, 22 (1983) 2335.
- (2) W.S. Bowers, T. Ohta, J.S. Cleere and P.A. Marsella, *Science*, 193 (1976) 542.

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The new world desert genera Encelia, Enceliopsis, Geraea and Fluorensia are members of a monophyletic group (here called Encelia group) within the tribe Heliantheae as indicated by joint morphological characters (1). These genera form also a major center of distribution of chromenes and benzofurans, a class of compounds common to several tribes of the Asteraceae (2). Phytochemical analyses revealed the presence of numerous novel compounds (3,4). The herbaceous genera Enceliopsis and Geraea yielded only benzofuran derivatives whereas the woody species of Encelia and Fluorensia elaborated both chromenes and benzofurans. Populational analyses proved the chemical patterns to be conservative and stable in regard to their composition. Quantitatively however as with Encelia farinosa significant differences were found along a west/east geographical gradient. Chemosystematic analyses of the chromene and benzofuran data indicated a closer affinity of the genera Encelia, Enceliopsis and Geraea compared to Fluorensia. Within Encelia three clades of species could be distinguished by their chromene and benzofuran patterns (5).

- (1) H. Robinson, Smiths. Contrb. Botany, 51, (1981) 48.
- (2) P. Proksch and E. Rodriguez, Phytochemistry, 22 (1983) 2335.
- (3) A. Mitsakos and P. Proksch, Biochem. Syst. Ecol., 13 (1985) 257.
- (4) P. Proksch, M. Proksch, W. Weck and E. Rodriguez, Z. Naturforsch. 40c, (1985) 301.
- (5) P. Proksch and C. Clark, Phytochemistry, in press.

Acknowledgements: Financial support of the DFG (Pr 229/1-1, 1-2) is gratefully acknowledged.

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Rhamnus lycioides L. (Rhamnaceae) is a spiny shrub growing in the brush-woods of the Mediterranean coastal regions of Spain. The aerial part is very frequently employed in folk medicine as a hypotensive and anti-glaucoma drug in the region of Valencia and it has never been described phytochemically. Studies on its hypotensive properties are being carried out with the most polar fractions which are pharmacologically active. However, middle polarity extract (EtOAc) showed a slight effect. The present study gives a report on the majority of the compounds (flavonoid aglycones) from that extract..

Plant material (leaves and stems), dried and powdered, was extracted with 70% MeOH and then successively fractionated with  $\text{CHCl}_3$ , EtOAc and BuOH. Flavonoid aglycones were detected in the  $\text{CHCl}_3$  and EtOAc extracts. By means of column chromatography (silicagel 60) and TLC (silicagel G-60 and cellulose) we have isolated eight substances; using UV-vis and  $^1\text{H}$ -NMR spectroscopy and TLC comparison with authentic samples, six flavonols have been identified: rhamnazin, rhamnocitrin, isorhamnetin, kaempferol, 3-OMe-quercetin (new in the Rhamnaceae family) and quercetin. In addition, we are in the process of elucidating the structure of two 5,7-dihydroxy-flavanones by  $^{13}\text{C}$ -NMR and MS techniques. This type of compounds is rather infrequent in the Rhamnaceae and was found previously in Rhamnus pallasii (1).

- (1) A. Sakushima, M. Coşkun, S. Hisada and S. Nishibe, *Phytochemistry*, 22 (1983) 1677.

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Die in den Moorgebieten Nordeuropas vorkommende Rosmarinheide wird gelegentlich als Verfälschung handelsüblicher Drogen beobachtet (1). Auch in neuester Literatur (2) wird diese Pflanze als giftig beschrieben, obwohl unsere früheren Untersuchungen (3) keine Anhaltspunkte für toxische Diterpene ergeben hatten.

Im Anschluß an diese Arbeiten konnten wir durch Tropfen-Gegenstrom-Chromatographie (DCCC) das Iridoid-glucosid Gardenosid und eine Reihe von Flavonolglycosiden isolieren. Letztere besteht aus den beiden isomeren Quercetinmonopentosiden Guaijaverin und Avicularin, sowie einem stark polaren Dipentosid, ebenfalls des Quercetins. Nach Aussage der  $^{13}\text{C}$ -NMR-spektroskopischen Daten handelt es sich hierbei um Quercetin-3-O-(2-1)- $\alpha$ -L-arabinofuranosido- $\beta$ -D-xylopyranosid.

Für diese bisher nicht beschriebene Verbindung schlagen wir den Namen Polifoliosid vor.

Auch diese erneuten Untersuchungen der Inhaltsstoffe von Andromeda polifolia geben keinen Hinweis auf die Giftigkeit der Pflanze.

- (1) Hagers Handbuch der Pharmazeutischen Praxis IV, 175  
Springer, Berlin/Heidelberg (1972)
- (2) Roth, L., Daunderer, M. und Kormann, K. "Giftpflanzen  
Pflanzengifte" Ecomed, München (1984)
- (3) Pachaly, P. et al. Arch. Pharm. 313 702 (1980)



COMPOSITION OF THE ESSENTIAL OIL OF OCIMUM CANUM GROWN  
IN RWANDA

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Ocimum canum Sims. occurs wild in tropical Africa as well as in other parts of the world. The species grows wild also in Rwanda (Central Africa), where it is used in traditional medicine and for aromatic purposes. In the literature much confusion exists about O. canum. The names O. canum, O. americanum and also O. basilicum and O. kili-mandscharicum have been interchanged, particularly for camphor-containing plants. Besides 'camphor-type', also 'methyl cinnamate-type', 'citral-type' and 'linalool-type' essential oils of O. canum have been reported. Because of the confusion about O. canum and since a taxonomic study on Ocimum species growing in Rwanda is performed at the moment, we were interested to investigate whether different chemical varieties of O. canum should be present in Rwanda.

Leaves and flowers of O. canum specimens growing wild in eastern and southern Rwanda (at Kibungo and Butare respectively) were subjected to hydrodistillation. The essential oil samples were analyzed by combined LSC and GLC, and by GC-MS. Comparison with authentic samples and mass spectrometry were used for identification of the essential oil components.

All essential oil samples were characterized by a high content of linalool (60-90%). Neither camphor nor citral and methyl cinnamate could be detected. The samples with the lowest content of linalool (ca. 60%) contained relatively large amounts of sesquiterpene hydrocarbons such as bergamotene (ca. 10%) and  $\beta$ -caryophyllene (ca. 5%). All monoterpene hydrocarbons were present in minor amounts, Timonene (ca. 1%) being the most important one. Oct-1-en-3-ol (ca. 2.5%) and 3-octanol (ca. 1.5%) were the only oxygen-containing components, besides linalool, which amounted to more than 1% in most of the oil samples.

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Hypericumzubereitungen erheben in der Volksmedizin u.a. den Anspruch bei Herz- und Kreislaufbeschwerden wirksam zu sein (1). Diese Indikation konnte bisher keiner Substanzklasse zugeordnet werden.

Neuerdings fand man Anhaltspunkte für die Anwesenheit von Procyanidinen (PC), die bei Crataegus für die Steigerung der Koronardurchblutung verantwortlich sind (2). Ziel unserer Arbeit war es, die genuine PC-Zusammensetzung zu ermitteln und zu untersuchen, ob Einzel-PC mit denen von Crataegus identisch sind. Dazu wurde ein alkoholischer Hypericum-Extrakt über Polyamid (Woelm) mit Ethanol (96%), Ethanol: DMF (8:2) und DMF in monomere Flavanderivate, nieder- und höher-oligomere PC aufgetrennt, oder an Sephadex-LH-20 (Pharmacia) mit Ethanol als Eluens fraktioniert.

Der Nachweis von Einzel-PC erfolgte mittels DC auf Kieselgelfertigplatten 60 F 254 (Merck) mit dem Fließmittel: Ethylacetat-Ameisensäure-Wasser-konz. Salzsäure-Cetrimid (85:6:8:1:0,5 OP) und Detektion mit Vanillin-Salzsäure, sowie durch HPLC auf einer RP 18 Säule. Die Konzentration der PC wurde photometrisch ermittelt (3). An Sephadex-LH-20 konnte ein PC isoliert werden, dessen massenspektroskopische und chromatographische Daten mit denen von PC B-2 aus Crataegus identisch sind. Der Gehalt von ca. 2% nieder-oligomeren PC im blühenden Johanniskraut und die Übereinstimmung dieser mit den PC aus Crataegus lassen vermuten, daß es sich bei den PC um das gesuchte Prinzip für die Herzwirksamkeit des Johanniskrautes handelt.

- (1) Tonero, A., Belg. Pat. 654.916 vom 16.02.1965,  
Appl. 27.10.1964 ref. C.A. 64, 19329 a (1966)
- (2) Weinges, K., Kloss, P., Trunzler, G. und Schuler C.:  
Planta med., Suppl. 4., 61 (1971)
- (3) Hölzl, J. und Strauch, A.: Planta med. 32, 141 (1977)

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Cachrys ferulacea (L.) Calestani (Prangos ferulacea Lindley) is a Mediterranean Umbellifer common in central and southern Italy. The plant is known for the big amount of forage it produces, the flavour which it yields to milk and cheese produced by cattle feeding on it (even the meat from cattle fed on the plant has a special aroma!) and for the edible mushroom Agaricus nebrodensis Inz. which grows on its roots (1). As the identity of the compounds responsible for the aroma is unknown, and moreover the taxonomic status of Cachrys ferulacea is still under discussion (2,3) and Italian populations have not been studied at all, we started a research into the plant with an investigation of anatomical characteristics and essential oil of the fruits.

Ripe fruits were collected from a population at 1400m on the Sibillini mountains (central Appennines). Fruit morphology and anatomy were observed with transmission-light and scanning-electron microscopy. Essential oil was collected by continuous hydrodistillation and studied by GC and GC-MS (polymethylsiloxane capillary columns).

Details of the microstructure of the fruit have been obtained. Main compounds in the essential oil are  $\alpha$ -pinene, sabinene,  $\beta$ -pinene, p-cymene, cis- and trans- $\beta$ -ocimene,  $\gamma$ -terpinene, trimethylbenzaldehyde, a monoterpenoid ester and a sesquiterpenoid hydrocarbon. The structure of the last two compounds has not been fully elucidated. Trimethylbenzaldehyde is supposed to be an artifact formed during the hydrodistillation from an (iso)ferulol-ester (4); indeed the compound is absent in an extract of the fruits. The structure of the ferulolester has not yet been determined. The presence of (iso)ferulol-esters in plants belonging to the tribus Smyrnieae has not been reported yet. The few, older, literature data on volatile compounds from Cachrys or Prangos species do not report on high boiling compounds.

The carpological information obtained is taxonomically relevant (3) and points to a tight relationship - if not identity - of Cachrys ferulacea from Italy to Prangos ferulacea described from Eastern habitats (3,5).

(1) A. Di Martino & F. M. Raimondo, Boll. Studi ed Inform. Giard. Col. Palermo 26 (1974) 116-129.

(2) T. G. Tutin, in Tutin, T. G. et al., Flora Europea vol. 2 (1968) 343.

(3) M. G. Pimenov & V. N. Tikhomirov, Feddes Repertorium 94 (1983) 145-164.

(4) K.-H. Kubeczka & I. Ullmann, Phytochem. 20 (1981) 828.

(5) I. Hernnstadt & C. C. Heyn, Boissiera 26 (1977) 1-91



SECRETORY STRUCTURES AND ESSENTIAL OIL COMPOSITION OF  
HERACLEUM SPHONDYLIIUM SUBSP.ORSINII LEAVES.

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Heracleum sphondylium L.s.l. is represented in Central Italy by the two subspecies H.sphondylium subsp.orsinii (Guss.) H.Neumayer and H.sphondylium subsp.ternatum (Velen.) Brummitt (according to Flora Europea). The occurrence of populations showing characteristics intermediate in some aspects between both subspecies has been reported (1). Preliminary reports dealt with the anatomy, morphology and volatile oil composition of different organs of plants from both subspecies (1,2).

In the frame of our research (3) into taxonomical and ecological relationships in Heracleum sphondylium s.l. a more thorough knowledge of the leaf oil composition of both taxa is needed. For obtaining leaf oil from subsp.orsinii we collected leaf material from two populations growing on Mount Terminillo (1700m) in the Central Appennines of Italy. One of the populations contained plants having ternate leaves (usually in addition to undivided leaves). In total five collections of leaves were made, one consisting only of ternate leaves. The oils were isolated by continuous hydrodistillation and studied by GC-FID, equipped with computing facilities, and by GC-MS. The oils consisted of a complex mixture (more than 150 detectable compounds) of mono- and sesquiterpenoid hydrocarbons and alcohols, aliphatic esters and phenylpropanoids. Most of the compounds were identified.

The main secretory structures of both subspecies proved to be secretory canals and glandular hairs. Secretory canals were present in pith, pericycle and cortex of the petiole, extending into the lamina beside the vascular bundles. Glandular hairs occurred along the veins of the lower leaf-side. In general the indumentum of the leaves proved to be quite characteristic: Subsp.orsinii leaves have a hispid indumentum, while subsp.ternatum leaves have a pubescent indumentum. The plants with ternate leaves from Mt. Terminillo have typical orsinii hairs.

Although a remarkable quantitative variation was observed, the qualitative composition of the five oils was identical. Statistical evaluation (BMDP statistical software package) gave no clearcut division as to population or leaf form. Oils from subsp.ternatum, from intermediate populations and of artificial hybrids are under study now; they have already been found to be quite different from orsinii oils.

(1) L.Montanarella, Thesis, Perugia 1984.

(2) M.R.Cagiotti, A.Menghini, L.Morelli, L.Montanarella. 2. Conv. Naz. S.I.F.

(3) T.A.van Cuijk, F.C.Fischer, M.M.V.Meeussen, R.Bos, G.Weimarck, [1984 Farm. Tschft. België 61 (1984) 401.

# HIGHLY METHOXYLATED FLAVONES OF SIDERITIS : A CHEMOTAXONOMIC SURVEY

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The species of the genus Sideritis (Lamiaceae) are well distributed in Mediterranean Spain and some of them have a great ethno-pharmacological significance, mainly as antiinflammatory drugs. The present work attempts to establish a relation between the methoxyflavone content and the systematic position of ten pure taxons of Sideritis growing in Eastern Spain. Aerial parts were extracted with hexane and chloroform. Organic extracts were treated with 11% Na<sub>2</sub>CO<sub>3</sub> and 4% NaOH. Flavonoid aglycones were isolated from alkaline fractions by preparative TLC on silicagel; solvents CHCl<sub>3</sub>-EtOAc (1:1) and C<sub>6</sub>H<sub>5</sub>Me-dioxane-AcOH (90:25:4). Identification was carried out by UV spectroscopy, according to VOIRIN rules(1), bidimensional TLC and HPLC(C-18, MeOH-H<sub>2</sub>O gradient elution)

Highly methoxylated flavones were not detected in the three taxons of the subsection Gymnocarpae F.Q.: S. incana incana, S. incana glauca and S. incana sericea. From the subsection Carpos-tegiatae F.Q. we have not found any flavone in S. scordioides but the other species did contain this type of compound. Three flavones: sideritoflavone, xanthomicrol and 8-methoxy-cirsilineol were identified in variable proportions in S. angustifolia, S. funkiana, S. leucantha, S. tragoriganum tragoriganum and Sideritis tragoriganum ssp nova. This last taxon is easily distinguishable from its standard species by its much higher 8-methoxy-cirsilineol content. S. javalambrensis was the species that showed the most singular flavone pattern; only gardenin D and xanthomicrol were detected in it. The flavone distribution is closely related to the species taxonomy, revealing the independence of S. scordioides and S. javalambrensis. The results are complementary and agree with those given in a recent paper about other Sideritis species(2).

(1) B. Voirin, *Phytochemistry*, 22 (1983) 2107

(2) F.A.T. Barber<sup>a</sup>n, J.M. N<sup>u</sup>ñez and F. Tom<sup>a</sup>s, *Phytochemistry*, 24 (1985) 1285

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Als wirksame Verbindungen vieler Drogen gelten lipophile, in Wasser unlösliche Verbindungen. Es stellt sich die Frage, ob und in welchen Konzentrationen diese potentiellen Wirkstoffe in die üblichen wässrigen Drogenzubereitungen gelangen. Diese Frage wurde geprüft am Beispiel Valeriana-officinalis-Radix (Mazerat, Infus: Valepotriate, Valerensäuren) (1), Curcuma-xanthorrhiza-Rhizom (Infus: Curcuminoiden) und Piper-methysticum-Rhizom (Mazerat: Pyrone) (2). Die Trennung und quantitative Bestimmung wurde mittels HPLC-Technik durchgeführt.

(1) R. Hänsel und J. Schulz, Pharm. Ind. 5, 531 (1985).

(2) J. Lazar, Dissertation, Berlin 1983.



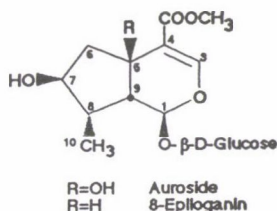
**Auroside, a new Iridoid Glucoside from *Phlomis aurea***M.F. Lahloub<sup>1</sup>, N. El-Sebakhy<sup>2</sup>, M. El-Ghazouly<sup>2</sup>G.-A. Gross<sup>3</sup>, O. Sticher<sup>3</sup>

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Due to the isolation of some new iridoid glucosides from *Phlomis fruticosa* [1-3], it was of interest to investigate the glycosidic constituents of various species belonging to the genus *Phlomis* (Labiateae) growing wild in Egypt, namely *P. aurea* Decna and *P. floccosa* D. Don. In the following we will describe the isolation and structure determination of a new iridoid glucoside from *Phlomis aurea*.



The dried plant material (whole plants) was extracted with methanol. The methanolic extract was fractionated by means of a series of chromatographic methods including silica gel, reversed phase and sephadex LH 20 column chromatography as well as preparative TLC giving a new iridoid glucoside, named auroside along with two acteoside type phenylpropanoid glycosides (acteoside and a not yet identified glycoside) and some other glycosides in trace quantities which contain an iridoid skeleton.

The structure of auroside was established by spectroscopic (UV, IR, FAB-MS, <sup>1</sup>H-NMR, and <sup>13</sup>C-NMR) means to be 5-hydroxy-8-epi-loganin.

**References:**[1] M.L. Scarpati, M. Guiso, Gazz. Chim. Ital. **99**, 1150 (1969).[2] A. Bianco, M. Guiso, C. Iavarone, C. Trogolo, Gazz. Chim. Ital. **105**, 185 (1975)[3] A. Bianco, C. Bonini, M. Guiso, C. Iavarone, C. Trogolo, Gazz. Chim. Ital. **107**, 67 (1977).

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Die zuerst von Herrmann (1) in mehreren Labiaten aufgefundene Rosmarinsäure ist, wie die vorliegenden Untersuchungen zeigen, ein geeigneter Leitstoff für zahlreiche Labiatendrogen und daraus hergestellte Fertigarzneimittel. Besonders reich an Rosmarinsäure sind die Melissenblätter mit Gehalten über 2% und insbesondere bestimmte *Melissa-officinalis*-Neuzüchtungen mit Gehalten über 3%. Vergleicht man die handelsüblichen Extrakte aus Labiatendrogen und die daraus hergestellten Fertigarzneimittel anhand des Leitstoffes Rosmarinsäure mit entsprechenden Standardextrakten, so entsprechen nur wenige Handelsextrakte und wenige Fertigarzneimittel den Erwartungswerten.

Zur Bestimmung der Rosmarinsäure wurde eine von Gracza (2) ausgearbeitete quantitative HPLC-Methode umgearbeitet und optimiert. Die neue Methode basiert auf der selektiven Anreicherung der Rosmarinsäure und Extraktion mit Wasser und anschließender Ausschüttelung mit Ether (s. Patent (3), gemäß DC-Prüfung einheitlich).

- (1) K. Herrmann, Arch. Pharm. 287, 142 (1954).
- (2) L. Gracza und P. Ruff, Arch. Pharm. 317, 339 (1984).
- (3) A. Nattermann und Cie. GmbH, Patent 3010040 (1980).

**Strukturaufklärung eines Apiose enthaltenden Iridoiddiglykosides mit Hilfe der H,H-verschiebungskorrelierten 2D-NMR-Spektroskopie**

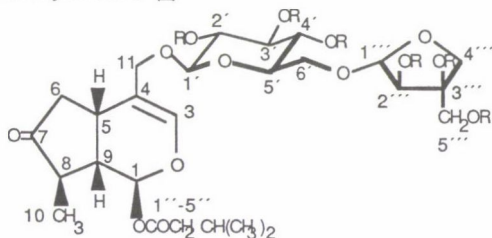
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Im Rahmen der Untersuchung des Methanolextraktes der Wurzeln von *Sambucus ebulus* L. (*Caprifoliaceae*) [1] gelang die Isolierung von 6'-O-β-D-Apiofuranosylebulosid (**1**)



1 R=H  
2 R=COCH<sub>3</sub>

Über die Konstitution des entsprechenden Monoglykosides (*Ebulosid*) berichteten wir bereits [2]<sup>\*)</sup>. Die Zuckerkomponente von **1** wurde durch hochauflösende <sup>1</sup>H-NMR-Spektroskopie unter Zuhilfenahme von H,H-COSY- Experimenten an **1** und **2** ermittelt. Damit stehen erstmals Hochfeld- <sup>1</sup>H-NMR-Daten von Apiose in diglykosidischer Bindung zur Verfügung. Die Zugehörigkeit der Apiose-Einheit zur D-Reihe der Monosaccharide folgt aus der Differenz der molekularen Drehungen von **1** und *Ebulosid* im Vergleich mit analogen Strukturen [3,4].

\*) Die Konfiguration der Methylgruppe C-10 musste nach weiterer Analyse von Hochfeldkernresonanzspektren aufgrund einer feststellbaren Kopplung von H-C(8) mit α-H-C(6) (W-Mechanismus) entgegen den in [2] gemachten Angaben für *Ebulosid* nach β korrigiert werden (siehe auch [1]).

In Vorzeichen und Grösse übereinstimmende COTTON - Effekte des Ketocarbonylchromophors von *Ebulosid* und 7-Dehydrologanin bekräftigen diesen Befund [5].

**Literatur:**

[1] G.A.Gross, Diss. ETH Zürich Nr. 7800, 1985.

[2] Farm. Tijdschr. Belg. 61(3), 233 (1984).

[3] T.Asahara, I.Sakakibara, T.Okuyama, S.Shibata, Planta med. 50(6), 488. (1984).

[4] R.K.Huyalkar, J.K.N.Jones, M.B.Perry, Can.J.Chem. 43, 2085 (1965).

[5] G.Snatzke, Private Mitteilung.



Strukturaufklärung eines neuen Typs dimerer Phenylpropanoidglykoside mit Hilfe der H,C-verschiebungskorrelierten 2D-NMR-Spektroskopie

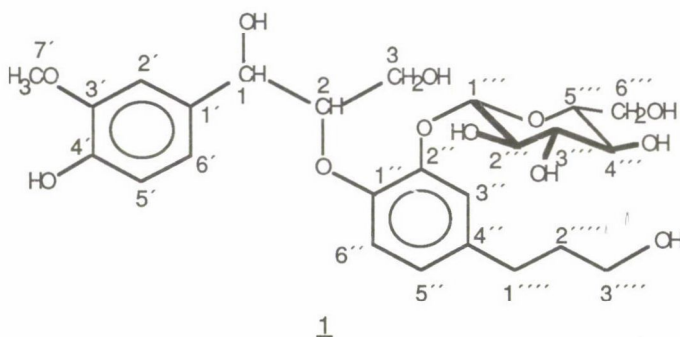
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Die systematische Aufarbeitung des Methanolextraktes der Wurzeln des Zwergholunders (*Sambucus ebulus* L.) [1] führte zur Isolierung von 1-(4-Hydroxy-3-methoxyphenyl)-2-O-(4-(3-hydroxypropyl)-2-β-D-glucopyranosyl)-phenyl)-glycerol (**1**), einem neuartigen dimeren Phenylpropanoidglykosid, das sich formal als Derivat von Glycerin ansehen lässt.



Seine Konstitution wurde mittels hochauflösender  $^1\text{H}$ - und  $^{13}\text{C}$ -NMR-Spektroskopie aufgeklärt. Die Ermittlung der aromatischen Substitution gelang durch Kombination der Analyse von NOE-Differenzspektren des Octaacetates von **1** mit H,C-COSY und H,C-COLOC-Experimenten [2,3] an **1**.

Literatur:

[1] G.A. Gross, Diss. ETH Zürich Nr.7800, 1985.

[2] H.Kessler, Ch.Griesinger, J.Lautz, Angew.Chemie **96**(6), 434 (1984).

[3] H.Kessler, Ch.Griesinger, J.Zarbock, H.R.Loosli, J.Mag.Res. **57**, 331 (1984).

COMPOSITION OF THE ESSENTIAL OIL IN LAMINA AND PETIOLE  
OF HERACLEUM DISSECTUM LEAVES.

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Heracleum dissectum is one of the H.sphondylium s.l.-related taxa of uncertain taxonomical position. Some authors consider it as a synonym of H.sphondylium subsp.montanum (Schleicher ex Gaudin) Briq.(1), while others consider it as a separate species, H.dissectum Ledeb. (2). In the frame of our studies on the taxonomy of H.sphondylium-related taxa (3) we grew plants from original seed material from Russia. Volatile oils from lamina and petiole were separately collected by continuous hydrodistillation. The composition of the oils was studied by GC and GC-MS (polymethylsiloxane capillary columns).

The oils consisted of a complex mixture (more than 100 detectable compounds) of mono- and sesquiterpenoid hydrocarbons and alcohols, aliphatic hydrocarbons, alcohols and esters, and phenylpropanoids. Most of the compounds could be identified, main compounds are: cis-hex-3-enol-1, ethylbutyrate, 2,4,6-octatriene,  $\alpha$ -thujene,  $\alpha$ -pinene, a menthatriene, limonene, cis- and trans  $\beta$ -ocimene,  $\gamma$ -terpinene, isobutylangelate, terpinolene, linalool, isoamyl-2-methylbutyrate, isoamylisovalerate, isoamylangelate, 4-terpineol, methylchavicol, terpenylacetate,  $\beta$ -bourbonene,  $\beta$ -elemene,  $\beta$ -caryophyllene, cadinol,  $\beta$ -humulene, phenylethylisovalerate,  $\gamma$ -cadinene,  $\delta$ -cadinene and nerolidol. The compositions of lamina and petiole oils were qualitatively the same, but the relative amounts of single compounds were remarkably different. The amount of oil in the petiole is, however, very low, causing the composition of the oil from complete leaves to be predominantly determined by the lamina oil. None the less a standardized sampling is necessary for obtaining the quantitative data necessary for statistical evaluation.

Finally the results were compared to previous (3) and unpublished (4) results on related taxa: H.dissectum proved to be related (as to the leaf oils) to part of the taxa united (1) in H.sphondylium subsp. montanum, viz. the taxa described as H.sphondylium subsp. granatense (Boiss.) Briq. and to the other mountainous taxa H.sphondylium subsp. pyrenaicum (Lam.) Bonnier & Layens and orsinii (Guss.) H. Neumayer.

(1) R.K. Brummitt, in Tutin, T.G. et al., Flora Europea, vol. 2, p. 364.

(2) I.P. Mandenova, in Komarov, V.I. et al., Flora of the USSR, vol. 16,

(3) T.A. van Cuijk, F.C. Fischer, M.M.V. Meeussen, R. Bos & G. Weimarck,  
Farm. Tschft. België 61 (1984) 401 <sup>p. 238</sup>

(4) F.C. Fischer, R. Bos, L. Montanarella & G. Weimarck, to be published.

69 | A Phytochemical Study of Vegetative Liquorice  
Plant..(Glycyrrhiza glabra L.)

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Inspite of the existence of several publications on the natural constituents of liquorice root, only very few reports have so far published about the steroidal and flavonoid constituents of the areal parts of the plant. So, it was deemed desirable to study the coumarinic, steroidal, terpenoid, flavonoid and flavonoid-glycoside constituents of such green parts of the plant to search for new compounds which may be of biological value. Fractionation of the flavonoid aglycones on preparative PC affords quercetin, m.p. 314-16° (0.49 %, dry weight basis); kaempferol, m.p. 279-80° (1.2 %) and methyl kaempferol as revealed by chromatographic colour reactions, UV data and bathochromic shifts induced by specific reagents. The flavonoid glycosides were resolved on polyamide column followed by successive PC to isolate kaempferol-3-O-glucoside. The position of the glucose moiety was confirmed by UV data and respective bathochromic shifts. The crude coumarin mixture was resolved by preparative alumina-chromatoplates followed by column chromatography of the separated zones on alumina to afford two furocoumarin compounds. These are, bergapten, m.p. 187-88° (0.0008%) and xanthotoxin, m.p. 142-43° (0.0032 %). Their identity was confirmed by UV, IR, NMR and MS spectral data. The existence of such two furocoumarins are encountered in liquorice plant for the first time. Surprisingly the areal part does not contain any terpenoid constituents. However,  $\beta$ -sitosterol was isolated from the unsaponifiable fraction (0.053 %).

✉ Alexander von-Humboldt Stipendiat from August 1  
till November 1 1985.



EFFECT OF DRYING CONDITIONS ON PRIMARY  
CARDENOLIDE CONTENT OF DIGITALIS LANATA  
ANALISED BY HPLC

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There are many contradictory publications /1-5/ about cardenolide composition of Digitalis lanata. As a result of analytical developments it can be established nowadays that living plants contain mainly primary glycosides and the amount of secondary glycosides increases depending on the processing of drug and analytical procedures.

Our investigations were carried out with D. lanata plants grown in phytotron and analysed by our HPLC method.

It has been established that secondary glycoside formation proceeds in different degree during drying of freshly harvested leaves at 20, 40, 60 and 80 °C. This decomposition of primary glycosides is caused by enzymes of D. lanata remaining active in the course of drying. Higher drying temperatures result in a mixed - primary and secondary cardenolides containing - drug because of the partial fermentative transformation. The dynamism of secondary glycosides formation has been established as a function of humidity loss in the course of drying.

The cardenolide composition of drug corresponding to the living conditions of leaves /plants/ can be reached and kept by drying under 40 °C temperature.

- /1/ Ligeti, G.: Pharmazie, 1957. 12:433
- /2/ Wolf, L., Bottyán, J., Karácsöny, E.M.: Planta Med., 1971. 20:36.
- /3/ Fischer, F., Bärtsch, H., Schmidt, H.J.: Pharmazie, 1971. 26:769.
- /4/ Pitra, J., Prochazka, V., Horák, P., Pötter, H., Bärtsch, H.: Pharmazie, 1976. 31:814.
- /5/ Wurst, Fr.: Arch. Pharm., 1983. 316:236.

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As a part of the systematic chemical and pharmacological investigation of *Sideritis* species growing in Spain, the essential oil from *S. mugronensis* x *angustifolia* was studied. Within species of this genus striking differences in the essential oil composition may exist, among other reasons because of the occurrence of chemical varieties and hybrids. We were interested in analyzing the essential oil isolated from this plant in order to establish its composition and evaluate the differences and similarities with the respective species type, *S. mugronensis* and *S. angustifolia*, whose qualitative and quantitative composition has been previously reported (1) (2).

Leaves and stems were subjected to steam distillation, yielding a yellowish essential oil that was fractionated on a silicagel column (using Hexane and Hexane-Dichloromethane mixtures) and then each fraction was analyzed by GLC and combined GC/MS. Comparison with authentic samples or with reported mass-spectra data were used for identification of the essential oil components (3).

A total of 49 components were identified of which 27 were the hydrocarbon compounds ( $\alpha$ -pinene, camphene,  $\beta$ -pinene, sabinene, myrcene,  $\alpha$ -phellandrene,  $\Delta^3$ -carene,  $\alpha$ -terpinene, limonene,  $\beta$ -phellandrene, p-cymene,  $\gamma$ -terpinene, terpinolene,  $\alpha$ -copaene,  $\beta$ -bourbonene,  $\beta$ -caryophyllene, alloaromadendrene,  $\alpha$ -humulene,  $\alpha$ -cubebene,  $\alpha$ -mourolene, germacrene-b,  $\alpha$ -curcumene,  $\beta$ -gurjunene,  $\delta$ -cadinene, calacorene, calamenene, calacorene isomer) and the other 22 belonged to the oxygenated fraction. These last compounds were: 1,8-cineole, linalool, cis-sabinene-hydrate, fenchone, thujanol, terpinen-4-ol, camphor, geranial, fenchyl acetate,  $\alpha$ -terpineole, myrtenal, bornyl acetate, isobornyl acetate, thymol,  $\alpha$ -terpinyl acetate, caryophyllene epoxide, three sesquiterpene alcohols, cadinol,  $\alpha$ -bisabolol, farnesol.

The oxygenated compound content (sesquiterpene alcohols) is higher, and  $\alpha$ -bisabolol is the main constituent of this essential oil.

The differences observed with the species type were only quantitative.

- (1) A. Villar, A. Navarro, M.C. Zafra-Polo and J.L. Rios, Pl. med. et phytother., 18 (1984) 150
- (2) C. Mateo, J. Sanz and J. Calderon, Phytochemistry, 23 (1984) 319
- (3) E. Stenhagen, S. Abrahamsson and F.W. McLafferty, TNO Zeist, 1979.

# BESTIMMUNG, DARSTELLUNG UND CHEMISCHE UMWANDLUNG VON CUMARINEN AUS CORTEX HIPPOCASTANI

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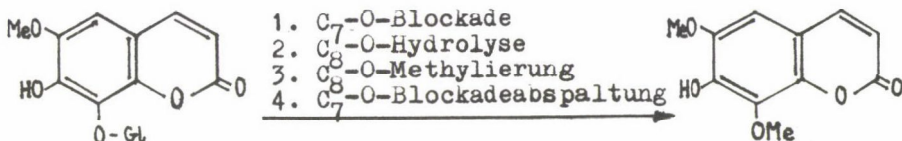
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Bei der Esculingewinnung aus Kastanienrinde fällt eine an Cumarinen reiche Fraktion als Nebenprodukt an. Neben einer gewissen Menge Restesculin enthält sie vor allem Fraxin und Scopolin /1/.

Es wurde eine Bestimmungsmethode entwickelt und damit der Gehalt der einzelnen Cumarine im Ausgangsmaterial und in den Verarbeitungsphasen kontrolliert /2/.

Durch Säulenchromatographie mit modifiziertem Polyamid konnten Esculin, Fraxin und Scopolin in 40-60% Ausbeute rein erhalten werden /3/.

Das auf diese Art angefallene, therapeutisch kaum interessante Fraxin wurde in einer 4-Stufensynthese in das medizinisch verwendbare Isofraxidin /4/ chemisch umgewandelt /5/.



Weitere Isofraxidinhomologa bzw. -derivate konnten auf gleiche Weise synthetisiert werden.

- /1/. Reppel, L. /1956/ Planta Med. 4, 199
- /2/. Gorecki, P., Mscisz, A. Herba Polon. /im Druck/
- /3/. Jerzmanowska, Z., Samuła, K. /1966/ Dissert. Pharm. Pharmacol. 18, 169
- /4/. Nieschulz, O., Schmiersahl, P. /1968/ Arzneim.-Forsch. 18, 1330
- /5/. Ahluwalia, V.K., Gupta, V.N., Rustagi, C.L., Seshadri, T.R. /1960/ J.Sci. Industr. Res. 19B, 345



SECOIRIDOIDGLYKOSIDE AUS FRÜCHTEN VON  
LIGUSTRUM VULGARE L.

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Unzureichende Kenntnisse über die Inhaltsstoffe der als toxisch geltenden Früchte von *Ligustrum vulgare* L. gaben Anlaß zu dieser Arbeit. Aus einem methanolischen Extrakt der reifen Früchte wurden säulenchromatographisch vier Secoiridoidglykoside in Mengen, die eine pharmakologische Prüfung ermöglichen, kristallin dargestellt. Diese wurden als Ligstrosid, Oleuropein und Nüzhenid sowie als LF(3)8, das in der Gattung *Ligustrum* bisher noch nicht gefunden wurde, mit den üblichen spektroskopischen Methoden identifiziert. LF(3)8 spaltet bei Methanolyse in Nüzhenid und Oleosid-7-methylester. Es ist folglich aus drei Glucose-, einer Tyrosol- und zwei Secoiridoid-(Oleosid-Aglucon)Einheiten zusammengesetzt. Vergleiche der physikalisch-chemischen Daten lassen den Schluß zu, daß LF(3)8 mit der von LaLonde et al. (1) aus den Samen von *Fraxinus americana* isolierten Verbindung Gl 3 identisch ist.

Die oben genannten Verbindungen treten mengenmäßig in der Reihenfolge LF(3)8 < Oleuropein < Ligstrosid < Nüzhenid auf.

- (1) R.T. LaLonde, Ch. Wong and A.I.-M. Tsai, *Journal of the American Chemical Society* (1976) 3007

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Das Wasser-Kreuzkraut (*Senecio aquaticus* HUDS.) ist in West- und Mitteleuropa stark verbreitet. Die Pflanze gedeiht vornehmlich auf feuchten Moorwiesen und in feuchten Gebüsch.

Bereits 1937 wurde *S. aquaticus* auf Pyrrolizidin-Alkaloide hin untersucht<sup>1)</sup>. Als Hauptalkaloid wurde das toxische Seneciphyllin isoliert.

1946 wurden Verfütterungsversuche durchgeführt und festgestellt, daß eine seit längerem beobachtete Viehkrankheit mit tödlichem Ausgang auf diese Pflanze zurückgeführt werden konnte<sup>2)</sup>.

Eine Varietät von *S. aquaticus* ist in Rumänien im nördlichen Karpatengebiet beheimatet. Es handelt sich um die *S. aquaticus* HUDS. var. *rosulatus* NYAR. die Pflanze ist deutlich kleiner als *aquaticus* und trägt im Gegensatz hierzu langgestielte Infloreszenzen. Sie kommt ebenfalls auf nassen Moorwiesen vor, wächst jedoch auch schon auf relativ feuchten Weidenflächen.

Da diese Varietät bislang auf Alkaloide hin noch nicht untersucht wurde, sollte eine phytochemische Bearbeitung sowohl chemotaxonomische Aspekte (Verwandtschaft mit *S. aquaticus*) liefern als auch einen Hinweis darauf geben, ob diese Varietät ebenfalls eine Gefährdung für Weidevieh darstellt.

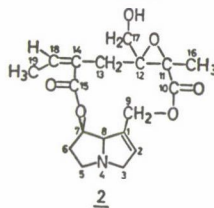
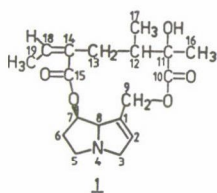
Die Pflanze wurde nördlich von Miercurea Ciuc gesammelt und auf Alkaloide hin untersucht. Dünnschichtchromatographisch konnten vier Alkaloide nachgewiesen werden, von denen zwei isoliert wurden. Es handelt sich dabei um das Hauptalkaloid Senecionin 1 und ein bislang noch nicht beschriebenes neues Pyrrolizidin-Alkaloid der Struktur 2.

Die Strukturen wurden durch spektroskopische Methoden ermittelt.

Aufgrund der aufgezeigten Strukturen sind beide Alkaloide als stark toxisch einzuschätzen.

1) J.J.Blacki, Pharm.J.138, 102 (1937)

2) W.C.Evans and E.T.R.Evans, Nature (London) 164, 30 (1949)



COUMARINS AND FLAVONOIDS FROM  
DAPHNE GNIDIoidES

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Since compounds like mezerein from Daphne mezereum and odoracin from D. odora exhibited antileukemic activity (1-2) we have initiated a study with Turkish species of Daphne to investigate them both chemically and pharmacologically . We have isolated known coumarins and flavonoids, daphnin, daphnetin, daphnetin 8-glucoside, esculin, daphnerotin, daphnetoxin, apigenin 7-glucoside, luteolin 7-glucoside, luteolin 4'-glucoside, quercetin 3-glucoside, isovitexin, vicianin 2 and triterpene  $\alpha$ -amyrin from the aerial parts of D. gnidioides. New acetyl-coumarins, 7-acetyl-umbelliferone, 7-acetyl-daphnetin, 8-acetyl-daphnetin, 8-acetyl-daphnin, 6-demethyl-7-acetyl-daphnerotin were also characterized from the same extract. The structures of the compounds were characterized by spectral methods. Although the crude extract showed a slight inhibition in the cell growth none of the isolated compounds consist the activity.

- (1) S.M. Kupchan and R.L. Baxter, Science, 187 (1975) 652.
- (2) S. Kogiso, K. Wada and K. Munakata, Agr. Biol. Chem. 40 (1976) 2119.



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Extracts obtained from the aerial parts of Phlomis lychnitis (Labiatae) are used in Spanish folk medicine as topical antiinflammatory drugs. In continuation with our studies on the flavonoid compounds from Labiatae species, we have now studied the flavonoids from this plant. The air-dried aerial parts were extracted with n-hexane,  $\text{CHCl}_3$  and EtOH in succession. The EtOH extract was paper chromatographed and several fractions which showed flavonoid colour when visualized under UV light were found. The principal fraction was studied. Its UV spectrum in MeOH (340i, 318, 300sh, 271, 254sh) showed the typical shape and values of a p-coumaroyl-glycoside (1). Mild alkaline hydrolysis yielded p-coumaric acid (TLC identified against authentic sample) and a flavone glycoside. The UV spectra in MeOH and after addition of the classical shift reagents (2), evidenced free hydroxyls at C-5 and C-4' and substituted hydroxyls at C-7 and C-3'. Acidic hydrolysis yielded chrysoeriol and glucose, and showed that the original glycoside was chrysoeriol-7-O-glucoside. This deacylated glucoside was permethylated and EIMS analysed. This analysis supported the existence of a chrysoeriol-7-O-glucoside ( $M^+$ , 546; A+H 328 m/z,  $T_1$  218 m/z) (3). The naturally occurring acylated glucoside was permethylated and EIMS analysed. The molecular ion was not observed, being the peak at m/z 532 (M-161) the first important ion, which corresponded to the loss of the acyl from the sugar moiety. An important fragment at m/z 161 (acyl moiety) was also observed. The position where the acid is linked to the sugar was evidenced by  $^1\text{H}$  NMR and the complete structure was chrysoeriol-7-O-beta-D-(3''-(E)-p-coumaroyl)glucoside, a new naturally occurring compound. p-Coumaroyl glucosides of luteolin and apigenin were also found, as well as the 7-O-glucosides of luteolin, apigenin and chrysoeriol.

- (1). F.R. Ansari, W.H. Ansari, W. Rahman, O. Seligmann, V.M. Chari and J. Osterdahl, *Planta Med.*, 36 (1979) 196.
- (2) T.J. Mabry, K.R. Markham and M.B. Thomas, *The Systematic Identification of Flavonoids*, Springer-Verlag, Heidelberg (1970).
- (3) H. Wagner and O. Seligmann, *Tetrahedron*, 29 (1973) 3029.

THE ESSENTIAL OIL FROM  
ELAEOSELINUM FOETIDUM (L.) BOISS. UMBELLIFERAE

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Elaeoselinum foetidum (L.) Boiss. is an endemic Umbellifera from the Iberian Peninsula whose essential oil is not yet known. As a great number of essential oils, are currently used in the pharmaceutical industry -either because of their aromatic properties or since, some of them, have defined pharmacological qualities-, we try to study those (unknown) with origin in Spanish plants.

The plants were distilled by steam distillation, and the yield was obtained using the Clevenger modified method. The percentages of essential oil obtained were the following: leaves 0,58% ; stems 0,53% and fruits 5%. Mature fruits were a richer source of oil than leaves or stems and, consequently, all subsequent work was carried on them. The results obtained are expressed as a percentage of total volatile oil in %/dry wt.

The physical data of the essential oil from mature fruits are:  $D_{20}^0$  0,85 ;  $n_D^{20}$  1,4713 and  $(\alpha)_D^{20}(\text{CHCl}_3)$  -15,29°.

The analysis of its chemical composition is performed by GC with capillary column and IR spectrometry.

32 components were detected in this same oil, 22 of which were identified by co-chromatography with authentic samples. Quantitative data is based on computer-calculated peak area without correction factors.

The essential oil was rich in monoterpene hydrocarbons. Major constituents were  $\alpha$ -pinene (77,3%) and  $\beta$ -pinene (17,3%) amounting to over 94% of the entire composition. Other constituents found were myrcene (1,16%), limonene (1,1%), sabinene (1%),  $\beta$ -phellandrene (0,4%), camphene (0,3%),  $\beta$ -caryophyllene (0,1%) and in lower concentration: linalol,  $\alpha$ -phellandrene, terpinen-4-ol, trans-ocymene, bornyl acetate, isoborneol,  $\gamma$ -terpinene, p-cymene, borneol,  $\alpha$ -terpineol, cis-ocymene, terpinolene,  $\alpha$ -terpinene and allo-ocymene.

COMPONENTS OF THE ESSENTIAL OIL OF  
ELAEOSELINUM TENUIFOLIUM (LAG.) LANGE. UMBELLIFERAE

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The fact that a great number of essential oils are currently used in the pharmaceutical industry -either because of their aromatic properties or since, some of them, have defined pharmacological qualities- induced us to study those, with origin y spanish plants, whose composition was not yet known. This is the case of Elaeoselinum tenuifolium (Lag.) Lange, Umbellifera with yellow flowers and which is endemic in the Iberian Península. (S., SE.)

The plants were distilled by steam distillation, and the yield was obtained using the Clevenger modified method. The oil was distilled from leaves, stems, flowers and fruits. It was discovered that the maximum content of oil was always found in mature fruits, and hence all subsequent work was carried on the mature fruits. The percentages obtained were: 4,63% in flowers; 0,66% in leaves; 0,49 % in stems and 4,93% in mature fruits. (% in dry wt.)

The physical data of the essential oil from mature fruits are:  $D_{20}^0$  0,832;  $n_D^{20}$  1,4777;  $(\alpha)_D^{20}(\text{CHCl}_3)$  -40,25°

The analysis of its chemical composition is performed by G.C. with capillary column and I.R. spectrometry.

24 components were detected, 15 of which were identified by comparing their retention time with those of authentic samples. Quantitative data are based on computer-calculated peaks area without correction factors.

The essential oil was rich in monoterpene hydrocarbons. The most abundant ones are myrcene (66,2%), sabine ne (17,2%), and limonene (8,7%), which amount to over 90% of the total composition. Other constituents found were  $\beta$ -phellandrene (1,6%), terpinen-4-ol (1,3%),  $\gamma$ -terpinene (0,7%),  $\alpha$ -pinene (0,6%),  $\beta$ -pinene (0,5%),  $\alpha$ -terpinene (0,3%), p-cymene (0,1%) and terpinolene (0,1%). And a lower concentration of linalyl acetate and canphene.



STUDY OF CASSIA ITALICA (MILL) F.W.ANDR. GROWING  
WILD IN IRAN

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Following is the result of study on Cassia italica we collected  
from Bandar-Abbas province (a port of Persian Gulf), this plant is  
growing wild in part of Iran.

The seeds of *C.italica* were cultivated in Tehran in 1100 m. altitude  
with the average temperature ranging from 28 - 40°C.

The comparison between above species and *Cassia angustifolia* was  
made for active constituents by spectrophotometric methods.

The active constituents of the above species is Sennosid B.

It contains 2.53-3.58 percent of Hydroxyanthracene derivative as  
Sennosid B, and it is comparable to Alexandria and Tinnevely  
Senna.

The phytochemical test was done on the leaves of the plant and  
as a result we found: Flavonoid, Saponin and Tannin.

The plantation was made in March and collection was made at the  
end August 1984.

- (1) A. Parsa, *Flore del Iran* 2, 482(1943)
- (2) E. Post, *Flora of Syria, Palestina, Sina*, I, 440(1932)
- (3) W. Pilarczyk, K.H.Ahrendt und M.Sesse, *Dtsch.Apoth-Ztg.* 116(1976)
- (4) E. Stahl, *Chromatographische und mikroskopische Analysen von  
Drogen*, P. 69(1970)

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Chemical investigations of the leaves of Ginkgo biloba L. (Ginkgoacées) led to the isolation and structural elucidation of five flavonol glycosides, and of a new unusual kaempferol derivative.

The leaves were exhausted with aqueous acetone. The aqueous acetone residue at first delipidated with diethyl ether, was extracted with ethyl acetate. The ethyl acetate extract was eluted on an anion-exchange resin column. The unadsorbed fraction gives the crude flavonoids extract which was refractionated successively on silica gel and polyamide columns.

The flavonoids were isolated and purified separately on sephadex columns. Their identity was proved by cristallisation, melting point, their colour reaction with different reagents, their maximum absorption and shifts obtained with diagnostic reagents, mass-spectroscopy and NMR-spectroscopy ( $^1\text{H}$  and  $^{13}\text{C}$ ).

The substances are : isorhamnetol-3-O-rutinoside (1), kaempferol-3-O-rutinoside (2), quercetol-3-O-glucoside (3), quercetol-3-O-rhamnoside and kaempferol-7-O-glucoside : these last two glycosides were identified in Ginkgo for the first time ;

kaempferol-3-O- $\alpha$ -(6"- p-coumaroyl-glycosyl- $\beta$ 1,4-rhamnoside) (4) : the new unusual molecule.

- (1) H. Geiger, and S. Beckman, Z. Naturfosch. (1965), 20 b, 1139
- (2) H. Geiger, Z. Naturforsch. (1979), 34 c, 878
- (3) K. Weinges, W. Bahr, P. Kloss, Arzneimittelforschung (1968), 18, 539
- (4) C. Nasr, M. Haag-Berrurier, A. Lobstein-Guth, R. Anton, in press

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Sanguisorba minor magnolii has a majority of flavonoid compounds on stems and leaves. Dry samples were ground down, defatted and extracted in the first place with ethylic ether in order to study free-aglycones, and secondly with 96% ethanol to study glycosides. Combined aglycones and sugars were studied using the acid hydrolysis plate method (2). Alcoholic extract drops were chromatographed (tlc cellulose, 60% acetic) and acid-hydrolysed on chromatographic plate in order to identify aglycones of each glycoside; aglycones were separated using the same chromatographic system in perpendicular way.

Aglycones were identified comparing with authentic markers. Aglycones characterization was confirmed by u. v. spectral measurements (3). Sugar fraction of isolated glycosides was studied by acid hydrolysis on plate and tlc on silicagel (1)(2).

All results are identical in stems and leaves. Free-aglycone were not detected in ethereal extracts. There are only two combined aglycones: Quercetin and Kaempferol.

Five glycosides were observed by chromatography in several solvents. They were identified as Quercetin-3-arabinoside, Kaempferol-3-arabinoside, Quercetin-3-glucoside, Kaempferol-3-glucoside and Kaempferol-3-glucourabinoside, by acid hydrolysis plate method and Rf-comparisons.

- (1) ANDARY, C., ROUSEL, J.L. RASCOL, J.P. PRIVAT, G. 1983. Colloque consacré aux Plantes Medicinales. Angers.
- (2) KARTING, T.H., MEGSCHAIDER, O. 1981. J. Chromat. 61, 375. 77.
- (3) MABRY, T.S. 1970. "The systematic identification of flavonoids". Ed. Springer-Verlag. New York.



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Die Blütenkörbchen von *Arnica montana* L. führen als Hauptwirkstoffe die Sesquiterpenlactone (=Sl) Helenalin und 11,13-Dihydrohelenalin sowie verschiedene Esterderivate dieser Verbindungen [1,2]. Eine Standardisierung der Droge und der daraus gewonnenen Zubereitungen sollte auf der Basis dieser Wirkstoffe erfolgen. Mit der von uns erarbeiteten quantitativen photometrischen und HPLC-Bestimmungsmethode [3] wurde das Ausmaß der quantitativen und qualitativen Variabilität in Arnikablüten untersucht.

Der Sl-Gesamtgehalt von bisher analysierten 26 verschiedenen Blütenherkünften lag im Bereich von 0,20-0,58%. In der Regel dominierten die Helenalinester, wobei das Verhältnis von Helenalinestern:11,13-Dihydrohelenalinestern im Bereich von 2:1 bis 5:1 lag (in Ausnahmefällen auch 9:1). Eine extrem andere qualitative Sl-Führung zeigte lediglich eine Blütendroge spanischer Herkunft, die praktisch nur 11,13-Dihydrohelenalinester führte. Mit Ausnahme dieser Drogenherkunft wurden qualitative Unterschiede in der Sl-Führung nicht gefunden. In den Zungen- und Röhrenblüten ist der Sl-Gehalt geringfügig höher als in den Hüllkelchblättern mit Blütenstandsboden, wobei letztere einen vergleichsweise höheren Anteil an 11,13-Dihydrohelenalinestern aufweisen.

Eine einjährige Lagerung der Blütendroge führte zu keiner qualitativen oder quantitativen Änderung in der Sl-Führung. In 15 Jahre lang gelagerter Droge wurde noch ein Sl-Gehalt von 0,10% gefunden.

[1] Willuhn, G., P.-M. Röttger, U. Matthiesen: *Planta Med.* 49, 226 (1983)

[2] Willuhn, G.: *Pharm. in uns. Zeit* 10, 1 (1981)

[3] Wandel, C., G. Willuhn: *Farm. Tijdschr. Belg.* 61e, 362 (1984)

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The phytoalexins produced by Phaseolus species due to their fungal infection have been reported by several authors (1-3). Most of the phytoalexins that have been isolated and characterized were found to be antifungal agents. A few of these compounds are known to be toxic for animals and possibly for man (4,5).

The present work deals with the study of phytoalexins produced by green bean pods (Phaseolus vulgaris L.) after inoculation with three fungi viz. Fusarium solani, Pencillium patulum and Phytophthora megasperma.

Five phytoalexins were isolated and identified as phaseollin (I) coumesterol (II), kievitone (III), phaseollidin (IV) and 6- $\alpha$ -hydroxyphaseollin (V). Their identity was proved according to m.p., TLC, UV and MS with comparison with authentics. Moreover, the preliminary screening of their antifungal activity were also carried.

#### References:

1. Biggs, D.R., J. Chem., 28, 1389 (1975).
2. Cruickshank, I.A.M., Biggs, D.R., Perrin, D. R. and Whittler, C.P., Phys. Plant pathol. (4), 261, (1974).
3. Van-Etten, H.D., Phytochem., (12), 1791, (1973).
4. Van-Etten, H.D., Phytopathol., 62, 795 (1972).
5. Wood, G.E., Advanced in Chemistry Series, No. 149 (1976).

ANTHRACENE DERIVATIVES FROM THE LEAVES OF PICRAMNIA  
PENTANDRA SW. (SIMAROUBACEAE)

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The leaves of Picramnia pentandra Sw. reputed in Haitian folk medicine as an antipyretic and as a gastric analgesic (3) has not been really investigated chemically until now. It was mentioned that 3-epibetulenolic acid (triterpenoid) was isolated from the bark (1) ; the seeds contain an unnamed alkaloid (1,2) and a large amount of fatty acids, but nothing was recorded in the literature concerning the chemical study of the leaves.

A preliminary study of the constituents of the leaves of this plant by thin-layer chromatography on the following extracts, cyclohexane, chloroform, ethyl acetate and methanol shows that this species is particularly rich in anthracene derivatives. Column chromatography and preparative HPLC were performed to isolate the main anthraquinone. Crystallisation in methanol, melting point and spectral datas by U.V., M.S. and <sup>1</sup>H-NMR were carried out to identify chrysophanol.

Furthermore, other anthracene derivatives are under investigation.

- (1) W. Herz, P.S. Santhanam and I. Wahlberg, *Phytochemistry*, 11, 3061-3063 (1972)
- (2) J.F. Morton, *Atlas of Medicinal Plants of Middle America*, Publ. C. Thomas (1981)
- (3) B. Weniger, Thèse de 3e cycle, *Toxicologie de l'Environnement*, Université de Metz, France (1985)



ON THE ISOLATION OF MACROCYCLIC DITERPENES  
FROM ROOTS OF JATROPHA GOSSYPIIFOLIA (Euphorbiaceae)

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Previously some antileukemic principles were isolated from the roots of *J. gossypiifolia* by Kupchan et al. (jatrophone (1)) and by Taylor et al. (2 $\alpha$ -hydroxy-jatrophone, 2 $\beta$ -hydroxyjatrophone, 2 $\beta$ -hydroxy-5,6-isojatrophone (2)). The jatropholones A and B, isolated by Purushothaman et al. (3), were inactive.

During the search for new antineoplastic compounds we isolated from *J. gossypiifolia* six macrocyclic diterpenes, the Jatropha factor J1 (jatrophone), Jatropha factors J2 and J3 (jatropholones A and B) and the new Jatropha factors J4 - J6. Jatropha factor J4 was identified as 3,4 $\alpha$ -epoxy-jatrophone. The factors J5 and J6 were characterized as 3,4 $\alpha$ - and 3,4 $\beta$ -epoxy-jatrophatrione. Jatropha factors J1, J2 and J3 are marginally irritant on the mouse ear. None of the Jatropha factors showed any significant antileukemic activity in the 3PS31 antileukemic system (lymphocytic leukemia P388).

- (1) S.M. Kupchan et al., J.Am.Chem.Soc. 92 (1970) 4476
- (2) M.D. Taylor et al., J.Am.Chem.Soc. 105 (1983) 3177
- (3) K.K. Purushothaman et al., Tetrahedron Lett. 11 (1979) 979

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Origanum majorana (L.) Family Labiatae is considered one of the most important aromatic plants and is widely distributed in Egypt. It was used in cookery as a spice and its volatile oil was used in flavour industry. It was also reported that Origanum compactum is used as abortifacient and for treatment of dysmenorrhea due to the high concentration of thymol and carvacrol in its essential oil (1). But no one has mentioned before anything about the presence of bitter glycosides in Origanum.

The powdered plant material of the total herb collected from the station of medicinal plants, Assiut University, Assiut (EGYPT), was firstly defatted with pet-ether. The defatted powder was successively extracted with ether, chloroform and lastly aqueous alcoholic extract. From the aqueous alcoholic extract, by use of column chromatography, silica gel "E" Merck (70-230) and ethyl acetate as eluant, we could isolate and identify phenolic glycosides arbutin and methylarbutin by means of their UV, IR, <sup>1</sup>H-NMR, <sup>13</sup>C-NMR and MS spectra in comparison with literature data. Small amounts of another two glycosides were also isolated and are under investigation. From the chloroformic extract, we could also isolate and identify hydroquinone and hydroquinone monomethylether.

Quantitative estimation of arbutin in Egyptian marjoram, adopting the spectrophotometric method used by Nguyen Hiep et al. (2) was found to be 1.44 g % of dried powdered herb.

We have found that hydroquinone has a potent cytotoxic activity on HTC hepatoma cells at 33 µg/ml.

Thus we can regard to the use of Egyptian marjoram herb as a good drug for the treatment of urinary tract infections (cystites, urethrites, and pyelitis) which refers to the presence of arbutin and methylarbutin and also as cytotoxic agent which refers to hydroquinone, besides its use as important source for essential oil.

1. C.O. Van der Broucke and J.A. Lemli, *Planta Medica*, 38, 317-331 (1980)
2. H. Nguyen, P.G. Delaveau et R.R. Paris, *Annales Pharm. Franç.*, 23, 5, 297-305 (1965)

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Production and utilization of essential oils and aromatic distilled waters has been a tradition in Iran since many years.

These distilled waters are named " ARAGHIYAT " by the native people and used as same as the medicinal plants.

The following plants are used for production of essential oils and aromatic distilled waters in many parts of Iran:

<u>Otostegia persica</u> Burm. ,	<u>Phlomis persica</u> Boiss. ,
<u>Zizyphora persica</u> Bge. ,	<u>Origanum viridis</u> (Boiss) Halas.
<u>Rosa canina</u> L. ,	<u>Thymus serpyllum</u> L. ,
<u>Fumaria parviflora</u> L. ,	<u>Cichorium Intybus</u> L. ,
<u>Mentha</u> spp. ,	<u>Cuminum cyminum</u> L. ,
<u>Foeniculum Vulgare</u> L. ,	<u>Carum carvi</u> L. ,
<u>Alhagi camelorum</u> Fisch. ,	<u>Pimpinella anisum</u> L.

- (1) Aynehchi-Salehi-Amin, Survey of Iranian Plants, Journal of Crud Drug Research, Vol. 19 , No. 2-3, 1981
- (2) Aynehchi-salehi-Amin, Survey of Iranian Plants, Journal of Crud Drug Research, Vol. 20 , No. 2, 1982



OCCURRING OF CALYCANTHINE : A BIS-TETRAHYDROQUINOLINE ALKALOID  
IN THE STEM-BARK OF PSYCHOTRIA FORSTERIANA A. GRAY (RUBIACEAE)

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Psychotria forsteriana A. Gray (Rubiaceae) occurs in a Pacific ocean island : VANUATU.

Previous works on the leaves showed that Psychotria forsteriana elaborates polyindoline alkaloids (1) that present some interesting pharmacological properties i.e. cytotoxicity, platelets aggregation (2) ...

The present communication is addressed to a simplified but highly selective extraction, isolation and identification of a non-poly-indolinic alkaloid from the stem-bark of this plant.

Air-dried stem-barks were collected by ORSTOM investigators in VANUATU (3). 2 kg were defatted with cyclohexane, then alkalinised with  $\text{NH}_4\text{OH}$  20 % and later extracted with chloroform in a soxhlet. The crude extract was acidified and washed with chloroform in order to remove various impurities. The aqueous acidic solution was realkalinised with  $\text{NH}_4\text{OH}$  20 % and extracted again with chloroform. The chloroformic extract was washed with water, dried on anhydrous  $\text{Na}_2\text{SO}_4$  and evaporated. The extract contained 3 major alkaloids and 5 minors (visualised per TLC revealed by ceric ammonium sulfate). One of the major alkaloids was separated by aluminium oxyde column chromatography, with toluene and various ratio of toluene- $\text{CHCl}_3$  mixtures. The alkaloid obtained in cristaline form (EtOH) was identified by comparison of its spectral data (UV, IR,  $^1\text{H-NMR}$ , MS, melting point, elementary analysis) with datas found in the literature (4), as calycanthine, a bis-tetrahydroquinoline alkaloid usually occurring in plants of the Calycanthaceae family. Calycanthine is a highly toxic substance that causes paralysis, violent convulsions and cardiac depression.

1. A. Roth, Contribution à l'étude de deux espèces du genre Psychotria  
Thèse de 3e cycle, Université Louis Pasteur, Strasbourg (1984)
2. A. Beretz, Polyindolinic alkaloids from Psychotria forsteriana :  
potent inhibitors of the aggregation of human platelets, under  
press (1985)
3. P. Cabalion, ORSTOM, Personal communications (1983)
4. N. Tatsuhiko, M. Alfonso, Planta Medica, 30, 186-188 (1976)

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The woodrotting fungus Inonotus obliquus (Pers. ex. Fr.) Pilat has for long been used as a folk medicine in some East European countries. The fungus is rich in  $\Delta^8$ -triterpenes of the lanosterol type and also contains  $\Delta^7$ -sterols, ergosterol and betulin. These compounds have been isolated and identified in order to carry out studies on the antitumour activity of these compounds (1).

Specimens of the fungus growing on birch trunks in nature and as laboratory cultures were investigated. The compounds were isolated from n-hexan extracts of the fungus by column chromatography. The structures were determined using IR, MS,  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR spectroscopy. GC-MS analyses were also used. The main compounds in the extract were inotodiol, lanosterol, trametenolic acid and  $3\beta$ -hydroxy-lanosta-8,24-dien-21-al (2-3). The characteristic fungal compounds,  $\Delta^7$ -sterols and ergosterol, occur only as minor constituents. The compounds stigmast-7-en- $3\beta$ -ol and betulin, found in these fungi, are not usually common among fungal products.

- (1) K. Kahlos et al., Farm.Tijdschr.Belg. 61 (3) (1984) 305
- (2) K. Kahlos et al., Planta Med. 50 (2) (1984) 197
- (3) K. Kahlos and R. Hiltunen, Acta Pharm.Fenn. 92 (1983) 220

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Das Kraut von Adonis vernalis L. weist etwa 30 Kedde-positive Verbindungen (2) auf, von denen heute 15 Cardenolide (3) bekannt sind. Zumeist handelt es sich um Monoglykoside, lediglich zwei Strophanthidin-Derivate weisen eine Zuckerkette auf.

Aus dem Extrakt von Herba Adonidis vernalis wurden durch kombinierte SC, DCCC und DC neben bereits beschriebenen Adonitoxigeninderivaten (4) drei weitere Cardenolide isoliert. Sie wurden als Adonitoxigenin-3-O- $\alpha$ -L-rhamnosido- $\beta$ -D-glucosid, Adonitoxigenin-3-[O- $\alpha$ -L(2'-O-Acetyl)rhamnosido-] $\beta$ -D-glucosid und Adonitoxigenin-3-[O- $\alpha$ -L(3'-O-Acetyl)rhamnosido-] $\beta$ -D-glucosid identifiziert. Die Strukturaufklärung erfolgte über dc-Nachweis von Aglykon und Zuckeranteilen nach hydrolytischer Spaltung auf der Kieselgelschicht (5), darüberhinaus mittels spektroskopischer Methoden (FD-MS,  $^1\text{H}$ -NMR,  $^{13}\text{C}$ -NMR).

Innerhalb der Gattung Adonis konnten damit erstmals Disaccharide des Adonitoxigenins nachgewiesen werden, die, wie die Strophanthidin-Derivate k-Strophanthosid und k-Strophanthin  $\beta$ , eine Zuckerkette mit endständig gebundener Glucose aufweisen.

(1) C. Winkler und M. Wichtl, *Planta med.* (im Druck)

(2) A. Pusz und S. Büchner, *Arzneim.-Forsch.*, 12(1962)932

(3) P. Junior und M. Wichtl, *Phytochemistry*, 19(1980)2193

(4) C. Winkler und M. Wichtl, *Pharm. Acta Helv.*, 60(1985)243

(5) D. Krüger und M. Wichtl, *Dtsch. Apoth. Ztg.*, 125(1985)55



## D. Flamme

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Bei der Beurteilung der therapeutischen Wirksamkeit von Crataegusextrakt-Präparaten zur Behandlung der beginnenden Herzleistungsschwäche nehmen die oligomeren Procyanidine (OPC) neben den kondensierbaren Flavanen und den Flavonoiden einen breiten Raum ein. Die Standardmonographie für Crataegus (4) läßt OPC als eine Stoffgruppe zur Beurteilung der Qualität von Arzneimitteln aus Crataegus-Früchten, -Blättern bzw. -Blüten zu. Ziel der Arbeit war es, ein möglichst genaues und selektives, aber auch schnelles Verfahren für die Routine-Qualitätskontrolle von Crataegus-Vor-, -Zwischen- und Endprodukten zu finden. Es wird ein Verfahren beschrieben, das es ermöglicht, oligomere Procyanidine des Weißdorns (Crataegus) aus der Droge und Extrakten bzw. Flüssigarzneimitteln selektiv abzutrennen, qualitativ und quantitativ zu bestimmen. In diesem Verfahren wird der Trockenextrakt an Sephadex LH 20 aufgereinigt, wobei die Trennung mittels DC (3) verfolgt wird. Folgende quantitativen Ergebnisse wurden gemessen:

a. Drogen (1 g je 50 ml Auszugsmittel Aceton/ Wasser 50 : 50 (V/V))	OPC-Gehalt, berechnet als (-)-Epicatechin	
	von Hersteller deklariert	gefunden
Fruct. Crataegi Fol. cum Flor. Crataegi		4,3 mg OPC/g 24,76 mg OPC/g
b. Fertigarzneimittel		
A. } B. } enthalten nur C. } Crataegus- D. } Auszüge	3 mg OPC/g ( $f_{20} = 0,9823$ ) 10 mg kondens. Flavane/ml 1,5 g Flavanderivate/100 ml berechnet als Leucocyanidol- biosid 75 g Preßsaft aus Cr. oxy- cantha/100 ml	1,20 mg OPC/ml 3,45 mg OPC/ml 3,04 mg OPC/ml 0,54 mg OPC/ml
E. } F. } Kombinationen mit anderen Inhaltsstoffen	4,00 g Tinct. Crataegi e fruct. recente/100 g 100 mg wässr.-ethanol. Fluid- extrakt aus gleichen Teilen Weißdornfrüchten, -blättern und -blüten 1:1/ml	0 mg OPC/ml 0,22 mg OPC/ml

\* Esbericard-Lösung, Ch.-Bez. 26173

Hersteller: Schaper & Brümmer, D-3320 Salzgitter 61 (Ringelheim)

3 H. Wagner, S. Bladt und E.M. Zgainski, Drogenanalyse, Springer-Verlag, Berlin, 1983, S. 304 ff.

4 Mitteilung des BGA vom 22.12.1983, veröffentlicht in BAnz. Nr. 1 vom 03.01.1984

Iola | ZUR KENNTNIS DER GERBSTOFFDROGEN—die  
Bestimmung der Gerbstoffe und adstringierende  
Wirkung

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Gerbstoffgehalt und adstringierende Wirkung wurde in frischen, ein Jahr alten oder mehr alten Drogen untersucht: *Qercus robur* L. (Cortex), *Hypericum perforatum* L. (Herba), *Rosmarinus officinalis* L. (Folium), *Fraxinus excelsior* L. (Cortex), *Fraxinus Oxycarpa* var. *pann.* Fukarek (Cortex), *Sambucus nigra* L. (Flos), *Centaurea minus* Moench. (Herba), *Thymus serpyllum* L. (Herba), *Primula vulgaris* Huds. (Folium).

Die Pflanzen wurden in Jugoslawien gesammelt. Die Gerbstoffbestimmungen wurden mit verschiedenen Methoden ermittelt: kolorimetrisch (1), gravimetrisch (2) und mit Casein (3).

Neben Gerbstoffe Wurde auch Gesamtpolyphenolgehalt untergesucht (3). Die Drogen wurden auch durch verschiedenen kolorimetrischen Reaktionen untergesucht und bei einigen Drogen wurde auch die Gerbstoffisolation durchgeführt (4).

Die adstringierende Wirkung untersuchte man mit Menschblut (5). Kolorimetrische Reaktionen mit den Drogenextrakten sowie auch mit isolierten Tanninen sind gut reproduzierbar und führen zu einer besseren Analytik der Drogen. Die Stärke der adstringierenden Wirkung ist in Korrelation mit den Tannin und Polyphenolgehalt. Das Altertum der Drogen führt zu einem langsamen Verringerung des Tanninhaltes.

- (1) Pharmacopoea Jugoslavica III, Beograd (1972).
- (2) Pharmacopoea Jugoslavica II, Beograd (1951).
- (3) Sohneider, G.: Arch. Pharm. 309 (1976) 38.
- (4) Bate-Smith, E.C.: Phytochemistry 20, (1981) 211.
- (5) Grujić-Vasić, J. et al.: Folia Med., 17 (1983) 89.

Andreas Bathe und Johannes Reisch

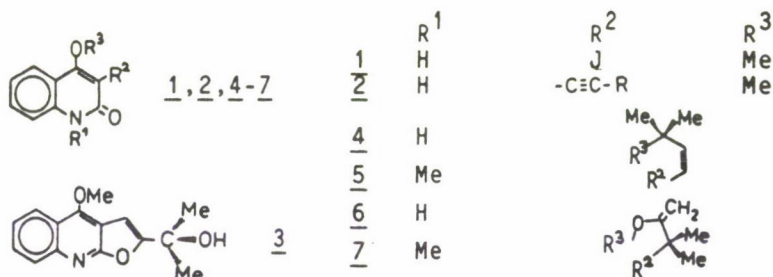
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Furo- und Pyranochinolone, Alkaloide diverser Rutaceen-  
gattungen, sind hinsichtlich ihrer biologischen Ak-  
tivität bislang kaum untersucht.

Der Einsatz der Stevens-Castro-Reaktion variiert nach  
Dieck/Heck<sup>1)</sup> ermöglichte nun in guten Ausbeuten ausgehen  
von 1 den Zugang zu Alkynylchinolonen des Typs 2 sowie  
zu linearen 4-Methoxy-Furochinolinen mit C-2-Substitutio-  
(z.B. 3). Die synthetische Breite, eine leichte Durchfüh-  
barkeit und die Markierungsmöglichkeit am Chinoyl-C-3  
sind Vorteile der neuen Sequenz.

Flindersin 4 und N-Methylflindersin 5<sup>+</sup>, einfachste  
Vertreter einer Reihe angulärer Pyranochinolone, konnte  
PT-katalysiert aus (N-Methyl)4-Hydroxy-chinolin-2-on und  
3-Chlor-3-methylbut-1-in gewonnen werden<sup>2)</sup>. Die gewünsch-  
te Dimethylpyrancyclisierung tritt über eine komplexe  
Reaktionskaskade ein, als Nebenprodukte fallen die Furo-  
chinoline 6 und 7 an, bei der Synthese von 5 zusätzlich  
ein Bischinoyl-Isopenten.

<sup>+</sup>) gemeinsam mit Dipl.-Chem. R.A. Salehi-Artimani



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1) B.A. Dieck, R.F. Heck, J. Organomet. Chem. 93, 259 (1975)

2) J. Reisch, A. Bathe und R.A. Salehi-Artimani, Arch.  
Pharm. (Weinheim) 1985, im Druck



# IRRITANT PRINCIPLES OF THE THYMELAEACEAE: RELATIONS OF STRUCTURES AND ANTINEOPLASTIC ACTIVITIES

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Some species of the Thymelaeaceae are known to exhibit irritant and tumor promoting (1-3) as well as antineoplastic activity (4). The principles which are responsible for these effects are polyfunctional derivatives of the diterpenes tigllane, daphnane or 1 $\alpha$ -alkyldaphnane a variety of which has been isolated from species of the genera *Daphne*, *Daphnopsis*, *Gnidia*, *Peddiea*, *Pimelea*, *Synaptolepis* and *Wikstroemia* monitored e.g. by the quantitative irritant assay on the mouse ear.

We now report on investigations to determine the antineoplastic activity of 17 polyfunctional diterpene esters of the daphnane and 1 $\alpha$ -alkyldaphnane type as revealed in the standard in vivo assay developed by the US National Cancer Institute (5) and established in our laboratory (3PS31, lymphocytic leukemia P388). Relations between structure and antileukemic activity as well as other biological activities of interest will be pointed out. Altogether the results demonstrate that most of the factors are more or less toxic in the doses required for significant antileukemic activity.

- (1) S. Zayed, W. Adolf, E. Hecker, *Planta Med.* 45 (1982) 67
- (2) W. Adolf, E. Hecker, *Planta Med.* 45 (1982) 177
- (3) A. Hafez, W. Adolf, E. Hecker, *Planta Med.* 49 (1983) 3
- (4) S.W. Kupchan et al., *J. Am. Chem. Soc.* 98 (1976) 5719
- (5) Geran, Greenberg et al., *Cancer Chemotherapy Rep.* Part 3, Vol.3, No.2 (1972) and Memoranda (pers.comm.)

R. Bauer, I. Khan und H. Wagner

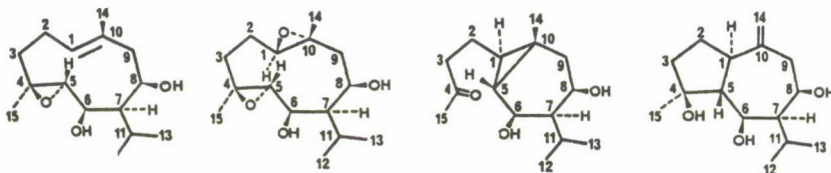
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In zahlreichen Präparaten zur unspezifischen Infektionsabwehr sind Extrakte aus Echinacea purpurea (L.) MOENCH. enthalten. Als mögliches Wirkprinzip wurde bisher eine Polysaccharidfraktion angesehen, die phagozytoseaktivierende Eigenschaften besitzt (1,2). Wir berichten nun über neue niedermolekulare Inhaltsstoffe, die ebenfalls an der Wirkung der Droge beteiligt sein könnten.

Aus den Wurzeln von E. purpurea wurden erstmals vier nichtflüchtige Sesquiterpenverbindungen isoliert und mit Röntgenstrukturanalyse und spektroskopischen Methoden identifiziert (3). Es handelt sich um die 8-O-Zimtsäureester von vier Sesquiterpenalkoholen mit Germacran- bzw. Guajan-Grundgerüst und den unten aufgeführten Strukturen 1 bis 4.

Als Analysenverfahren wird neben der Kieselgel-DC ein C18-Umkehrphasen-HPLC-System angegeben, das für die Standardisierung der Droge und zur Unterscheidung von E. angustifolia verwendet werden kann (4). Der damit bestimmte Gesamtgehalt der Ester beträgt ca. 1,0%.

Erste pharmakologische Tests der Ester lassen einen Einfluß auf das unspezifische Immunsystem vermuten.



Echinadiol(1) Epoxyechinadiol(2) Echinaxanthol(3) Dihydroxynardol(4)

- (1) H.Wagner, A.Proksch, I.Riess-Maurer, A.Vollmar, S.Odenthal, H.Stuppner, K.Jurcic, M.LeTurdu und Y.H.Heur, *Arzneim.-Forsch.* 34 (1984) 659.
- (2) M.Stimpel, A.Proksch, H.Wagner und M.-L.Lohmann-Matthes, *Infection and Immunity*, 46 (1984) 845.
- (3) R.Bauer, I.Khan, H.Lotter, V.Wray und H.Wagner, *Helv. Chim. Acta*, im Druck.
- (4) R.Bauer, I.Khan und H.Wagner, Publikation in Vorbereitung.

ANREICHERUNG UND CHARAKTERISIERUNG IMMUNOLOGISCH AKTIVER INHALTSSTOFFE AUS ECHINACEAE ANGUSTIFOLIAE RADIX UND ECHINACEAE PURPUREAE RADIX

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Eine Reihe von Mikroorganismen und Pflanzen enthalten Mono- bzw. Polysaccharide, die in-vitro und in-vivo eine immunmodulierende Wirkung besitzen (1). Die wegen ihrer immunstimulatorischen Wirkung schon seit langem bei grippalen Infekten verwendeten Asteraceen Echinacea purpurea (L.) Moench und Echinacea angustifolia DC enthalten als Wirkstoffe Heteropolysaccharide (2). Aus dem oberirdischen Teil von Echinacea purpurea wurden zwei immunstimulierende Polysaccharide rein dargestellt und strukturell aufgeklärt (3). Ziel unserer Untersuchungen war es, immunologisch aktive Inhaltsstoffe aus den Wurzeln beider Echinacea-Spezies anzureichern bzw. zu isolieren. Für die Anreicherungs-schritte wurden Diafiltrationen mit Hollow-Fiber Patronen und Säulenchromatographien an Sephadex Gelen eingesetzt. Die so gewonnenen Wirkfraktionen wurden in verschiedenen immunologischen Testsystemen eingesetzt. Die Aktivierung von Maus-Lymphozyten durch Drogeninhaltsstoffe wurde mittels Messung des  $6\text{-}^3\text{H}$ -Thymidin Einbaus in die DNS nachgewiesen (Mitogenstimulation). Die Stimulation der B-Lymphozyten wurde im Jerne-Plaque Test geprüft. Bei den hochmolekularen Polysacchariden aus Echinacea angustifolia und Echinacea purpurea konnte in-vitro eine starke mitogene Aktivität festgestellt werden. Nach unseren bisherigen Untersuchungen handelt es sich um B-Zellmitogene, wie durch Messung des  $[6\text{-}^3\text{H}]$ -Thymidineinbaus und der Antikörperbildung gezeigt werden konnte. In bezug auf Aktivität und Spezifität der isolierten Polysaccharide konnten in den verwendeten Testsystemen keine Unterschiede zwischen Echinacea purpurea und Echinacea angustifolia festgestellt werden.

(1) Lindequist, U. und Teuscher, E., Pharmazie 40 (1985) 10

(2) Becker, H., DAZ, 122 (1982) 2321

(3) Proksch, A., Diss. München (1982)



# HYPOTENSIVE ACTIVITY FROM SOME MEDITERRANEAN MEDICINAL PLANTS

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The hypotensive activity of Sideritis angustifolia Lag., Crucianella maritima L. and Rhamnus lycioides L., three medicinal plants used in the Mediterranean area of Spain, was studied.

The lyophilized aqueous extracts were assayed in groups of seven urethane anesthetized Wistar rats weighing 200-300 g. The samples were dissolved in physiological saline and were administered (0.25 ml) through a venous canula with the aid of an additional injection of saline (0.1 ml). Blood pressure was recorded via a pressure transducer on a polygraph (1). The hypotensive activity was compared with that of dihydralazin.

Plant material most active pharmacologically (R.lycioides L.) was further extracted with 70% MeOH, which was concentrated under reduced pressure to yield the extract which was fractionated with water-AcOEt and water-BuOH. All these extracts and the aqueous marc were pharmacologically tested. After observing the results of the activity, we then selected the BuOH extract which was further chromatographed on a column. Studies are continuing in order to identify the active principle involved.

	dose	% red. systolic pressure at 5'	% red. diastolic pressure at 5'
Dihydralazin	1 mg/Kg	16.09 **	41.26 *
<u>S.angustifolia</u>	150 mg/Kg	0.56 ns	- 1.10 ns
<u>C.maritima</u>	150 mg/Kg	2.43 ns	- 2.04 ns
<u>R.lycioides</u> :			
aqueous ext.	150 mg/Kg	25.00 *	32.30 *
aqueous ext.	50 mg/Kg	10.53 *	25.40 **
70% MeOH ext.	150 mg/Kg	24.21 *	37.50 *
AcOEt ext.	50 mg/Kg	10.43 *	24.39 **
BuOH ext.	50 mg/Kg	25.20 **	21.80 *
aqueous marc	50 mg/Kg	0.61 ns	11.50 ns

ns =not significant    \*P < 0.05    \*\*P < 0.01

- (1) J. Genest, O. Kuchel, P. Hamet and M. Cantin, Hipertension, Mc Graw-Hill Company, London, (1983), p. 1262.

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No qualitative variation in the alkaloidal content of the different organs of *Fumaria parviflora* Lam. was observed, although they varied quantitatively when assayed by non-aqueous titration. After column chromatography of extract, protopine and another alkaloid, suggested to be the desmethyl congener of the synthetic tetrahydroescholamine were isolated. The two alkaloids were characterized by their m. ps., chromatographic behaviour as well as UV, IR, NMR and mass spectra. Toxicological and pharmacological investigation of the ethanolic extract of *F. parviflora* as well as the alkaloid "protopine" isolated thereof were carried out. Both exhibited a significant decrease in intestinal muscle contractions.

Significant cardioinhibitory, antiarrhythmic, hypotensive and antipyretic effects were also found. The ethanolic extract of the plant as well as protopine can, thus be used as intestinal antispasmodic, antihypertensive and antiarrhythmic.

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Earlier investigation of Poke root (Phytolacca americana) resulted in the isolation of an active principle (1), which was later on identified as phytolaccatoxin (2), whose physiological activity was directly correlated in all respects to that reported for the roots. Some other reports on the isolation of moluscicidal saponin (semmatoxin) from P. dodecandra (3) and an anti-inflammatory triterpenoid (jaligonic acid) from P. esculenta (4), have been reported. Several studies concerning the isolation and structure elucidation of the saponin constituents of phytolacca species were also reported.

The saponin fraction of Phytolacca americana was found to possess high spermicidal activity (5). Clinical application of the active principle (saponin fraction), on 100 fertile cases eager for contraception, then followed up for one year revealed that no conception took place (5).

It was recommended as an effective local contraceptive drug.

The present work deals with the isolation of the active principle from the plant roots, which was successfully cultivated in Egypt.

An improved technic for the method of its separation and purification was performed. Moreover, the determination of the mutagenic capacity of the isolated spermicidal principle by means of the Ames test was carried out. The results obtained did not show any mutagenic capacity for the used concentrations.

#### References:

- (1) Ahmed Z.F., Zufall C.J. and Jenkins C.L.: J.Am. Pharm. A.soc., 38, 8(1949).
- (2) George H.S., Bernard M.M. and Virginia F.S.: J.Am.Chem.Soc. 86 (5) 957 (1964)
- (3) Parkharst R.M., Thomas D.W., Skinner W.A. and Carry L.W.: Can. J. Chem. 52 (5), 702 (1974)
- (4) Woo Won Sik, Lloydia, 36 (3), 326 (1973)
- (5) Ahmed Z.F. and Shaaban A.H.: Gaz. Egypt Soc. Gynaecol. Obstet. 9, 27, (1959).



108 | ARTEMISIA BIOASSAY IN DETERMINING THE ACUTE  
TOXICITY OF USNIC ACID AND SOME LICHENS

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The brine shrimp, Artemia salina Leach, has lately been used in pharmacological studies (1 and 2). In this work the brine shrimp bioassay was used to determine the short term toxicity of (-)-usnic acid and some lichen samples.

The LC<sub>50</sub>-24 hr for usnic acid was 5.5 µg/ml. The extracts of 34 Cladina and Cladonia species were also tested using this method (5 mg dry lichen sample/5 ml 0.1 % NaHCO<sub>3</sub>). The mortality rate of the brine shrimps was for 10 of the samples, 50-100 %. All these samples contained more than 0.5 % usnic acid. The amount of usnic acid in the lichen samples was determined by the reversed-phase HPLC method (3). Six samples containing no usnic acid gave a mortality rate of 5-60 %. 18 samples were inactive in the bioassay. These arbitrarily selected lichen samples represent about 10 % of the known Cladina and Cladonia species.

It can be concluded that the most toxic compound in these lichen samples is probably usnic acid, which is used for medicinal purposes because of its antibacterial effect.

- (1) B. N. Meyer et al., *Planta Medica* 45 (1982) 31-34
- (2) R. T. Trotter et al., *Journal of Ethnopharmacology* 8 (1983) 113-119
- (3) K. E. Huovinen et al., *Planta Medica* 45 (1982) 152

110 | IN VITRO CYTOTOXICITY OF SOME SESQUITERPENE LACTONES ON A  
HUMAN LUNG CARCINOMA CELL LINE

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In this in vitro study cytotoxicity of some sesquiterpene lactones (germacranolides) from *Eupatorium cannabinum* L. (Asteraceae) and related compounds have been tested on a human small-cell lung carcinoma cell line (GLC-4: Groningen Lung Carcinoma), using the Fast Green dye exclusion Assay, as described by Weisenthal et al. (1), with slight modifications.

The sesquiterpene lactones from *Eupatorium cannabinum* L. (eupatoriopicrin, eupatoriopicrin acetone, "substance 1" and hiyodorilactone E) showed highest cytotoxicity (ID 50 1-2 µg/ml) following 1 h incubation.

Moieties of the molecule (eupatolide, α-methylene γ-butyrolactone, angelic and tiglic acid) and related eudesmanolides (alantolactone, isoalantolactone) were less or not active in the test.

From the results it can be concluded that:

- a) the whole germacranolide sesquiterpene lactone molecule is necessary for optimal cytotoxicity;
- b) the in vitro cytotoxic activity of the sesquiterpene lactones increases with decreasing hydrophilicity;
- c) the germacranolides tested are more cytotoxic than the eudesmanolides.

- (1) L.M. Weisenthal, J.A. Marsden, P.L. Dill, C.K. Macaluso, Cancer Research, 43 (1983) 749-757.

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Chamomile (*Matricaria recutita* L.) extracts and compounds have been used medicinally because of their anti-inflammatory and papaverine-like spasmolytic activities. Most interesting compounds among these have been the lipophilic compound, (-)- $\alpha$ -bisabolol and the hydrophilic compounds, apigenin and apigenin-7-glucoside(1). Owing to the fact that they exhibit spasmolytic activity it was decided to test their calcium antagonism on vascular smooth muscle in vitro.

The lipophilic hydrodistilled volatile oil of chamomile was analysed by gas-liquid chromatography (GC) and the hydrophilic methanol extract of chamomile by high performance liquid chromatography (HPLC).

Calcium antagonism was tested by a method based on the depolarization-induced contraction of rabbit aortic rings. The contractions were evoked by 55 mM KCl in the presence of 2.5 mM  $\text{CaCl}_2$  in Krebs-Henseleit buffer solution (2).

According to the test (-)- $\alpha$ -bisabolol (0.15 % solution in 10 ml organ bath) caused a marked irreversible inhibition of the KCl contraction. It also evoked some contraction of the smooth muscle preparation itself. The volatile oil (contains 3% (-)- $\alpha$ -bisabolol) itself inhibited the KCl contraction over a similar concentration range as (-)- $\alpha$ -bisabolol. Such potent spasmolytic compounds as apigenin and apigenin-7-glucoside were, however, inactive according to this test. A high concentration of methanol extract of chamomile (contains 0.08 % apigenin and 2.47 % apigenin-7-glucoside) showed similar activity to (-)- $\alpha$ -bisabolol and the volatile oil.

The present study shows that those lipophilic compounds which have spasmolytic activity may also act as calcium antagonists. However, spasmolytic compounds such as apigenin and its glucoside showed no antagonistic activity according to this test. The activity of the methanol extract may be connected to the presence of volatile oil compounds in it.

1. U. Achterrath-Tuckermann et al., *Planta Medica*, 39 (1980) 38

2. R. Hof and H. Vuorela, *J. Pharmacol. Methods*, 9 (1983) 41



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*Santolina chamaecyparissus* L. is a species of the Asteraceae family that grows in eastern Spain. The flower of this plant is used in folk medicine because of its antispasmodic, digestive, antiinflammatory, antiseptic and antimicrobial properties (1).

The fresh flowers were distilled in a Clevenger apparatus, and the chemical composition of the essential oil were studied by GLC and GC-MS after fractionation on a chromatography column (silica gel and hexane/dichloromethane).

Antimicrobial activity was carried out by the agar dilution method (2). The essential oil was tested against 3 grampositive bacteria, 3 gramnegative bacteria and one yeast.

A total of 50 components were detected of which 39 were identified:  $\alpha$ -pinene, camphene,  $\beta$ -pinene, sabinene, myrcene,  $\alpha$ -phellandrene,  $\Delta^3$ -carene,  $\alpha$ -terpinene, limonene,  $\beta$ -phellandrene, p-cymene, 1,8-cineole,  $\gamma$ -terpinene, terpinolene, artemisia ketone, p-cymenene, fenchone, thujone, cis-sabinene-hydrate, borneol, camphor, terpinen-4-ol,  $\alpha$ -terpineol, myrtenal, bornyl acetate, isobornyl acetate,  $\alpha$ -cubebene,  $\alpha$ -copaene, 4-isopropyl benzaldehyde,  $\alpha$ -ylangene,  $\beta$ -gurjunene, carvacrol,  $\alpha$ -gurjunene, isocaryophyllene, allo-aromadendrene,  $\alpha$ -mourolene, geracrene-b,  $\delta$ -cadinene and cadinol. Two sesquiterpene alcohols were detected ( $M^+$  = 222-220) but they have not been identified. Nine other components have been detected but they have not been characterized. Camphor (25.19%), p-cymene, 1,8-cineole, allo-aromadendrene and  $\alpha$ -mourolene are the main constituents, totalling 61.50% of the oil.

The essential oil from *S. chamaecyparissus* has activity against grampositive bacteria and the yeast assayed but it has no activity against gramnegative bacteria. The growth of *Bacillus subtilis* has been inhibited at 1/400, *Staphylococcus aureus* at 1/400 and *Mycobacterium phlei* at 1/2500, *Candida albicans* is not grow at 1/800 dilution.

- (1) P. Font Quer, Plantas Medicinales. El Dioscórides renovado. 8<sup>a</sup> ed. Labor, Barcelona (1983)
- (2) L.A. Mitscher, R.P. Leu, M.S. Bathala, W.N. Wu, J.L. Beal and R. White, Lloydia, 35 (1972) 157.

113 | 1'-ACETOXYCHAVICOL ACETATE, A FUNGITOXIC COMPOUND FROM  
THE RHIZOMES OF ALPINIA GALANGA

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Alpinia galanga (Zingiberaceae) is native to south-eastern Asia. The rhizomes of the plant are widely used as a spice, called laos, and in traditional medicine, among others to cure skin infections caused by fungi. The essential oils isolated from fresh and dried rhizomes of A. galanga showed an antimicrobial activity against gram-positive bacteria, a yeast and some dermatophytes, using the agar overlay technique. The main components of the oils were also tested, and terpinen-4-ol was found most active.

An n-pentane/diethyl ether extract of dried rhizomes was active against Trichophyton mentagrophytes. Fractions showing activity were purified by column chromatography and TLC. Spectroscopic methods (MS, NMR) were used for the identification of the active component that proved to be 1'-acetoxychavicol acetate. Other components such as 1'-acetoxyeugenol acetate and 1'-hydroxychavicol acetate showed no activity.

Acetoxychavicol acetate was active against the seven fungi tested and its MIC value for dermatophytes ranged from 50 to 250 mg/l; dried sliced rhizomes contained 1.5% of this compound. The compound was not found in rhizomes of Alpinia officinarum, Zingiber officinale and Kaempferia galanga.

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In der Absicht, unter den erfahrungsmedizinisch verwendeten Drogen mit nachgesagter Herzwirksamkeit (1) Reinglykoside zu finden und darzustellen, wurden alkoholische Extrakte aus Corchorus capsularis-Samen hergestellt. Die biologische Wirkwertuntersuchung dieser Auszüge am Meerschweinchen ergab neben typischen Veränderungen der Chronotropie und Inotropie einen DL 100 Wert von 1,42 mg/kg i. V.. Die chromatographische Untersuchung von zwei über RLCC-Chromatographie abgetrennten Extraktfraktionen zeigte, daß sich diese aus drei Hauptcardenoliden zusammensetzen. Ihre Reindarstellung durch präp. HPLC und anschließende Strukturaufklärung führte zur strukturellen Übereinstimmung mit Strophantidin, Corchorosid A und Olitorosid [Gluco(1 → 4)Corchorosid A]. Parallel mit den Reinigungsschritten ließ sich die Glykosidwirkungssteigerung im Bioassay nachweisen. Olitorosid und Corchorosid A unterscheiden sich demnach in ihrer pharmakologischen Wirkung in Untersuchungen am spontan schlagenden rechten bzw. gereizten linken Meerschweinchenvorhof nur geringfügig. Konzentrationsabhängig wurde eine ausgeprägte Inotropiesteigerung für Corchorosid A bzw. Olitorosid angetroffen. Das Maximum der Kontraktionskraftzunahme lag für Corchorosid A bei ca. 300 % bei einer Konzentration von 2,5 µg/ml und für Olitorosid bei ca. 180 % bei einer Konzentration von 5 µg/ml. Beide Dosen führen bei entsprechender Einwirkzeit zu Intoxikationen des Meerschweinchenvorhofes. Aufgrund der analytischen Daten und des biologischen Wirkvergleiches ist anzunehmen, daß die untersuchten Reinglykoside Hauptwirkungsträger der Samenextrakte sind. Ein weiterer literaturbeschriebener polarer Inhaltsstoff mit Cardenolidstruktur (2) wurde wegen seines geringen Vorkommens in den von uns hergestellten Samenextraktfraktionen nicht näher in weitere Untersuchungen mit einbezogen.

- (1) Hagers Handbuch der Pharm. Praxis, Bd. 4, Springer Verlag, Berlin, Heidelberg, New York, S. 298 - 299
- (2) G.S. Ricca, C. Casagrande, Gazz. Chim. Ital. 112 (1982) 349 - 352.



115 | EFFECT OF SOME SUPPLEMENT DIETARY-FAT OILS ON THE FATTY  
ACID COMPOSITION OF PLASMA LIPIDS

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As most of the vegetable oils used for human nutrition have a very low content of  $\omega 3$  fatty acids, the inclusion of marine lipids has become an important means of balancing the diet. Furthermore, eicosa-pentaenoic acid (20:5 $\omega 3$ ;EPA) has been found to afford protection against thrombosis (1). On the other hand, gammalinolenic acid (18:3 $\omega 6$ ;GLA) is special among  $\omega 6$  vegetable fatty acids in that it has a number of beneficial properties (2). The aim of the present study was to investigate the effects of fat supplementation on plasma fatty acid composition.

The lipids were extracted as described earlier (3) and the fatty acids were determined by GLC using a PTV (programmed temperature vaporizer) sampling technique on a DANI HR 3800 PTV GC fitted with an OV-351 vitreous silica column (4).

The mean precision for the GC equipment was 1.7% (C.V.), and the extraction, derivatization and GLC together had a mean precision of 2.0% (C.V.) for 15 compounds.

The level of arachidonic acid (20:4 $\omega 6$ ;AA), which was 30% higher in the Finnish control group (N=43) than that for the Eskimos (N=32), was significantly reduced by consuming cod-liver oil at doses 2-15g/d. The EPA concentration, which in the Finnish control group was only 1/3 that of the Eskimos, increased significantly following a dose of 15g cod-liver oil/d. This was also the case following an intake of 3-4g EPA concentrate. After this dietary supplementation, the EPA level did not differ significantly from that for the Eskimos (3.2%). Neither wheatkernel oil or lecitin were found to have any significant effect on the AA or EPA levels at daily doses as high as 10-15g. On the other hand, a daily dose of 4g of oil from evening primrose seeds containing GLA significantly decreased the AA content.

The present study indicates that cod-liver oil, as well as EPA concentrates, can be used for raising low EPA levels in plasma lipids. GLA and rather small doses of cod-liver oil are also useful supplements having a decreasing effect on arachidonic acid level.

- (1) Dyerberg et al., Lancet, July (1978) 117.
- (2) Horrobin, D.F. (ed.), Clinical Uses of Essential Fatty Acids, Eden Press, Montreal, pp. 214 (1982).
- (3) Folch et al., J.Biol.Chem. 226 (1957) 497.
- (4) Laakso et al., Farm.Tijdschr.Belg. 61 (3) (1984) 373.

116 | COMPARATIVE STUDY OF ANTIMICROBIAL ACTIVITY  
AND CHEMICAL COMPOSITION OF SIDERITIS ESSENTIAL OILS

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Essential oils from medicinal plants are used in treating numerous disorders. A genus of Lamiaceae -Sideritis- is employed in Spain against various diseases. In previous work we have investigated the antimicrobial activity of different extracts of Sideritis species (1). In this work we have studied the antimicrobial activity of some essential oils from Sideritis species and their relation with the chemical composition. Volatile oils are used in treatment of infectious diseases, principally against grampositive bacteria.

Essential oils were obtained by steam distillation and their composition was studied by GLC. Antimicrobial activity screening was carried out by the agar dilution method (2). The essential oils were tested for activity against some bacteria and a yeast.

The species analyzed were :S. angustifolia Lag., S. funkiana Willk., S. javalambrensis Pau, S. leucantha Cav., S. mugronensis Borja and S. tragoriganum Lag. nova.

The principal compounds of this essential oils are :S. angustifolia (cadinol,  $\delta$ -cadinene and unknown sesquiterpene alcohols); S. funkiana ( $\beta$ -phellandrene and unknown sesquiterpene alcohols); S. javalambrensis ( $\alpha$ -pinene and sabinene); S. leucantha ( $\alpha$ -pinene, sabinene, fenchone); S. mugronensis (1,8-cineole, caryophyllene,  $\delta$ -cadinene); S. tragoriganum nova ( $\delta$ -cadinene, bisabolol,  $\beta$ -gurjunene and unknown sesquiterpene alcohols).

Only grampositive bacteria and yeast are inhibited by essential oil from Sideritis species and they have no activity against gramnegative bacilli. Essential oils of S. angustifolia and S. tragoriganum nova inhibited the growth of Mycobacterium phlei at a dilution of 1/800 and S. mugronensis at 1/400. Candida albicans does not grow when there is essential oil from S. angustifolia at 1/800 in the medium or from S. funkiana at 1/400. The antimicrobial activity against S. aureus is of no interest (1/100 and 1/200).

- (1) A. Villar, J.L. Rios, M.C. Zafra-Polo, M. Mares, M.C. Recio, *Plantes Médicinales et phytothérapie* (in press).
- (2) L.A. Mitscher, R.P. Leu, M.S. Bathala, W-N Wu, J.L. Beal, R. White, *Lloydia*, 35 (1972) 157.

IN VITRO IMMUNOMODULATION BY SRI LANKAN PLANTS.  
PART 2. EFFECTS IN THE MIGRATION INHIBITION FACTOR (MIF)  
TEST.

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Immunomodulation might contribute to activities exerted by certain Sri Lankan plants, used in the traditional Ayurveda system of medicine as tonics, vitalizers, roborants, stimulants, rejuvenatives etc. (1). The site of these actions can include humoral components like the complement system (2), and/or cellular components of the immune system.

A number of Sri Lankan plants, which are therapeutically used as mentioned above, has been investigated for their effects on the MIF, an in vitro correlate for cell-mediated immunity (3) using human lymphocytes as source of lymphokine and the murine myelomonocytic leukemia cell line WEHI<sub>3</sub> as target cells (4,5).

Some of the studied plant extracts show significant suppression of migration inhibition, very probably due to a suppression of MIF-production by the lymphocytes.

The results of this study seem to support the view that immunomodulation might indeed be at least partly responsible for the stimulating properties claimed for the investigated Sri Lankan plants.

The stem bark extract of *Azadirachta indica* A. Juss. (Meliaceae), being one of the most active samples, will be subjected to further phytochemical and immunological investigations.

1. R.P. Labadie, Ned. Tijdschr. Integr. Geneesk., 2 (1985), in press
2. See Part 1 of these series: Effects on the activation of human complement
3. I. Roitt, in: Essential Immunology, 4th Ed., Blackwell Scientific Publications, Oxford, (1980), p. 243
4. M.C.M. Kersten, E. Pels, R.A. de Weger and W. den Otter, J. Immunol. Methods, 33 (1980) 387
5. J.M. van der Nat, J.H. Beijnen, W.J.M. Underberg and R.P. Labadie, submitted for publication.



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Anthracyclines are a group of antineoplastic agents of natural origin (1). Their clinical use is limited by a dose-dependent cardiotoxicity. Also side effects concerning the immune system are reported frequently.

The search for less toxic derivatives yielded a series of (semi-) synthetic analogues. Though the immunomodulating effects of the best-known anthracyclines doxorubicin and daunorubicin have been thoroughly investigated (2) little has been published on the newer analogues. For this reason it seemed interesting to compare the effects of some of the newer anthracyclines to those of doxorubicin and daunorubicin in our in vitro test systems for humoral and cell-mediated immunity (complement activation and Migration Inhibition Factor (=MIF) tests respectively).

On both the classical and alternative pathways of activation of human complement, as measured in the haemolytic micro assay procedure (3) neither of the investigated compounds show a significant effect.

However in the MIF test using human lymphocytes as source of lymphokine and murine WEHI<sub>3</sub> monocytes as target cells (4,5) most of the anthracyclines cause a significant suppression of migration inhibition. The extent of the immunosuppressive activity appears to be proportional to the lipophilicity of the compound in question, expressed as HPLC retention coefficients.

1. F. Arcamone, in: Topics in Antibiotic Chemistry, P.G. Sammes (Ed.), Vol. 2, E. Horwood Ltd., (1978) 99
2. A. Mantovani, N. Polentarutti, W. Luini, G. Peri and F. Spreafico, J. Natl. Cancer Inst. 63 (1979) 61 and references cited
3. J.P.A.M. Klerx, C.J. Beukelman, H. van Dijk and J.M.N. Willers, J. Immunol. Methods 33, (1980) 215
4. M.C.M. Kersten, E. Pels, R.A. de Weger and W. den Otter, J. Immunol. Methods, 33 (1980) 387
5. J.M. van der Nat, J.H. Beijnen, W.J.M. Underberg and R.P. Labadie, submitted for publication.

IN VITRO IMMUNOMODULATION BY SRI LANKAN PLANTS.  
PART 1. EFFECTS ON THE ACTIVATION OF HUMAN COMPLEMENT

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The mode of action of a variety of Sri Lankan plants, used in the traditional Ayurveda system of medicine as tonics, vitalizers, roborants, stimulants and rejuvenatives is not yet understood. There is growing evidence that some influence on the human immune system, especially on the non-specific components, might be involved in such activities (1,2). In this respect, measuring of effects on the activation of the complement system is a very useful technique to study influences on non-specific humoral immunity (3).

A number of Sri Lankan plants with claimed activities as mentioned above has been investigated for effects on both classical and alternative pathways of activation of human complement, using the haemolytic assay procedure (4) modified to a microscale (5).

Some of the studied plant extracts show significant inhibition of both classical and alternative pathways of activation of human complement while others inhibit only one of the two pathways. The latter selectivity might be of mechanistic and practical interest.

The results of this study seem to support the view that immunomodulation might indeed be at least partly responsible for the stimulating properties traditionally claimed for the investigated Sri Lankan plants.

The stem bark extract of Azadirachta indica A. Juss. (Meliaceae), being the most active sample, will be subjected to further phytochemical and immunological investigations.

1. R.P. Labadie, Ned. Tijdschr. Integr. Geneesk., 2 (1985), in press
2. A. Bamunuarachchi, A. Abeysekera, K.T.D. de Silva and R.P. Labadie, Pharm. Weekbl. 119, (1984)
3. J.H. van Meer, Pharm. Weekbl., 119 (1984) 836
4. H. van Dijk, P.M. Rademaker and J.M.N. Willers, J. Immunol. Methods 36, 39 (1980) 29, 257
5. J.P.A.M. Klerx, C.J. Beukelman, H. van Dijk and J.M.N. Willers, J. Immunol. Methods 63 (1983) 215

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In verschiedenen Aristolochia-Arten wurden neben Aristolochiasäure I (ASI) und II (ASII) auch einige ihrer Reduktionsprodukte wie Aristolactam I und II (cyclisches Reduktionsprodukt) oder Aristolsäure I (Nitrogruppen-freie ASI) und die Desmethylverbindungen Aristolochiasäure Ia und Aristolactam Ia aufgefunden (1,2,3).

ASI und ASII wurden in der Humanmedizin u.a. zur Immunstimulation therapeutisch eingesetzt. Da sich bei chronischen Toxizitätsstudien an Ratten zeigte, daß diese Verbindungen zu Tumoren führen können (4), wurden 1981 sämtliche Aristolochiasäure-haltigen Arzneimittel aus dem Verkehr gezogen. Im Zusammenhang mit diesen Toxizitätsstudien wurden Untersuchungen zur Verstoffwechselung von ASI und ASII durchgeführt. Nach oraler Gabe konnten diese beiden Verbindungen im Urin der Ratte nicht mehr nachgewiesen werden, sondern nur noch eine Reihe von Metaboliten, die teilweise im Gegensatz zu den Muttersubstanzen eine intensive Fluoreszenz zeigten. Ihre Reindarstellung aus dem Urin erfolgte durch Chromatographie an Amberlite XAD-2, Kieselgel und Sephadex LH-20. Die Strukturauklärung gelang mittels  $^1\text{H}$ -NMR-Spektroskopie und Massenspektrometrie sowie durch Partialsynthese. Bei ASI waren Aristolochiasäure Ia und Aristolactam Ia Hauptmetaboliten, Aristolactam I und Aristolsäure I Nebenmetaboliten. Im Falle von ASII wurden Aristolactam II und Aristolactam Ia (Hydroxylierungsprodukt von Aristolactam II) als Hauptmetaboliten isoliert. Aristolactam I und II wurden auch beim Kaninchen und Hund als Metaboliten nachgewiesen. Somit sind die genannten Verbindungen nicht nur Pflanzeninhaltsstoffe, sondern werden auch im tierischen Organismus gebildet. Zusätzlich wurden jedoch Metaboliten (z.B. entmethylierte Aristolsäure I oder Nitrogruppen-freie ASII), die aus Pflanzen bisher noch nicht isoliert worden sind, nachgewiesen.

- (1) D.B. Mix, H. Guinaudeau und M. Shamma, J. Nat. Prod. 45 (1982) 657
- (2) S.C. Pakrashi, P. Gosh-Dastidar, S. Basu und B. Achari, Phytochemistry 16 (1977) 1103
- (3) H.A. Priestap, Phytochemistry 24 (1985) 849
- (4) U. Mengs, W. Lang und J.-A. Poch, Arch. Toxicol. 51 (1982) 107



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In *Styla Maydis* konnten neben den darin bereits bekannten Flavonoidglykosiden Maysin, 3'-Deoximaysin und 3'-Methylmaysin (1) erstmals Vitexin nachgewiesen werden. Die Identifizierung der mittels SC, präparativer PC und DC isolierten Flavonoide erfolgte durch Cochromatographie, UV- und NMR-Spektroskopie (2).

Vergleichende Untersuchungen mittels DC (3) und HPLC (1) über die Flavonoidmuster und -gehalte der *Styla* von 25 verschiedenen Maissorten erbrachten folgende Ergebnisse: Zwischen den Flavonoidgehalten der Griffel und den Korn-typen (Zahn- oder Hartmais bzw. Mischtypen) bestehen Zusammenhänge. Doppelhybridsorten weisen wesentlich höhere Flavonoidgehalte auf als Einfach- oder Dreiwegkreuzungen. Der Flavonoidgehalt der *Styla* ist zur Zeit der Pollenreife am höchsten und nimmt bis zur Fruchtreife kontinuierlich ab. Trocknungsversuche zeigten u.a., daß bei der Luft-trocknung der Flavonoidgehalt sich nur wenig ändert.

In orientierenden Untersuchungen wurden die diuretische und saluretische Wirkung des Infuses aus *Styla Maydis* an Ratten geprüft. Als Vergleich dienten reines Wasser und eine Lösung von Harnstoff in Wasser. Die Ergebnisse wurden mit dem zweiseitigen t-Test nach STUDENT (4) ausgewertet. Während die diuretische Wirkung nicht besonders stark ausgeprägt war, zeigte sich eine starke Erhöhung der Salurese, die in Beziehung zum Flavonoidgehalt der Droge zu stehen scheint.

- (1) ELLIGER, C.A., CHAN, B.G., WAISS, A.C., LUNDIN, R.E. and HADDON, W.F.: *Phytochemistry* 19, 293 (1980)
- (2) MABRY, T.J., MARKHAM, K.R., THOMAS, M.B.: *The Systematic Identification of Flavonoids*, Springer-Verlag, New York (1970)
- (3) STAHL, E.: *Dünnschichtchromatographie*, pp. 657-671, Springer-Verlag, Berlin-Heidelberg-New York (1967)
- (4) CAVALLI-SFORZA, L.: *"Biometrie. Grundzüge biologisch-medizinischer Statistik"*, G. Fischer Verlag, Stuttgart, 1974

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Acute intraperitoneal toxicity studies as well as phytochemical investigation for different plant extracts were carried out. Isolation of alkaloids is reported as a first record in this species. Quantitative seasonal variations of flavonoids and coumarins of Sonchus oleraceus at different stages for plants collected from different localities is discussed.

ISOLATION OF SILYMARIN FROM SILYBUM MARIANUM (L.) GAERTH  
GROWING IN EGYPT.

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The present work aims mainly to adopt a simple technique for the active principle silymarin from the seeds of either the plant growing wild or the recently cultivated, which might be liable to be applied for mass production of the product.

Trials to attain and improved new technique for the isolation of silymarin were successfully approached. The method of isolation comprises the use successive solvent fractionation. Quantitative evaluation of the isolated products was carried out by HPLC in comparison with authentic standards as well as some pharmaceutical drugs namely legalon and dura silymarin. The results revealed that the isolated samples were very pure and contained silibinin (=silybin A + silybin B) and silychristin in higher levels. Their values are much more higher than those of legalon. However silydianin is only present in a small amount, which may be due to different chemical strain of Silybium marianum as the starting material.

Moreover, several experiments for the cultivation of the plant (growing wild), in different localities especially the newly reclaimed lands in Egypt were carried out successfully.



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Tropic hormones, like TSH and the gonadotropins, react with plant extracts and several oxidated phenolic plant constituents under impairment or complete loss of their biological activities.

In own investigations the mechanism of interaction was scrutinized via CD(circular dichroism)-spectroscopy, the loss of activity could be correlated to changes in the secondary structure of the respective hormones.

In this study influences on the hormone-substrate-reaction, especially the structural influences of the phenolic inhibitor should be investigated. Commercially available hormones (Thyroid Stimulating Hormone, Pregnant Mare Serum Gonadotropin) were purified by exclusion chromatography. Fractions of high molecular weight showing good stimulating activity (TSH1, PMSG1) were taken for further investigations. Hormone fractions were incubated together with phenolic substrates in buffered solution and chromatographed on Sephadex G 100 to separate the hormone-substrate-complex from excessive substrate. The UV-spectra of the complexes give information on the amount of substrate bound, since the molecular absorbance coefficient of the phenolics clearly exceeds the one of the proteohormone.

Changes in secondary structure of the hormone were registered via changes in intensity of their CD-spectra. In addition, the structural composition of the hormone, regarding the share of  $\alpha$ -,  $\beta$ - and random-conformation, was determined by a computerized fit. Binding of graded doses of *Lycopus europaeus* as well as verbascoside to TSH1 shows as clear dose-dependency. Saturation limits were found at 1.5 mg, for the plant extract and 300  $\mu$ mol for verbascoside against 0.25 IU TSH. This correlates properly to impairments of stimulating activity of these hormone-substrate-complexes.

A complex from PMSG with oxidated caffeic acid, isolated by means of gel chromatography, proved to be biologically inactive as well. It's CD-activity was significantly augmented, compared to PMSG itself.

Unoxidated plant acids like chlorogenic acid or acids with blocked phenolic groups like ferulic acid showed no binding to tropic hormones, which is in agreement with their complete lack in antihormonal activity. Thus unblocked hydroxyls, which are reactily oxidated and polymerized seem to be essential for the binding and inhibition of the hormone.

# INFLUENCE OF THE NATURAL FLAVONOID HYPOLAETIN-8-GLUCOSIDE ON CAPILLARY PERMEABILITY

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Hypolaetin-8-glucoside (H-8-G) isolated from *Sideritis mugro-nensis* Borja (Labiatae) (1) and possessing anti-inflammatory and anti-ulcer activity in rats (2) has been studied to determine its influence on capillary permeability and isolated rat uterus contractions induced by several mediators of inflammation.

I- Capillary permeability: increased by histamine according to Tarayre et al. (3) on female Wistar rats, being administered H-8-G and troxerutin subcutaneously.

H-8-G was more active than troxerutin, a flavonoid derivative that reduce capillary fragility and permeability, in decreasing histamine-induced capillary permeability.

H-8-G			TROXERUTIN			mg/kg (s.c.) % Inhibition
100	200	300	200	300	400	
34'6*	42'8*	64'2**	24'6	32'7*	35'5*	

(\*  $p < 0.05$ , \*\*  $p < 0.01$  Student's t-test)

II- Isolated rat uterus contractions: the influence of H-8-G ( $2.5 \cdot 10^{-5}$  -  $2.5 \cdot 10^{-7}$  M) was determined using cumulative dose-response curves (4) of serotonin and  $PGE_2$  and single-dose curves of bradykinin. H-8-G did not exert any antagonism in the isolated rat uterus against serotonin,  $PGE_2$  and bradykinin.

The higher activity of H-8-G on capillary permeability as compared with troxerutin, a well-known drug used in the treatment of haemorrhoids and venous disorders of the lower limbs, might account for the therapeutic potential of the former compound.

- (1) A. Villar et al., *Planta Med.*, 1985, 70
- (2) A. Villar et al., *J. Pharm. Pharmacol.*, 36, 1984, 820
- (3) J.P. Tarayre et al., *Ann. pharm. franc.*, 33, 1975, 467
- (4) J.M. Van Rossum, *Arch. Int. Pharmacodyn.*, 143, 1963, 290

ISOLATION AND BIOLOGICAL ACTIVITIES OF NAPHTHOQUINONES  
AND SAPONINS FROM DIOSPYROS ZOMBENSIS (EBENACEAE)

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Diospyros zombensis is a tree from Malaŵi (Southeast Africa) which is phytochemically investigated here for the first time. By flash and Lobar chromatography of the ligroin and chloroform extracts of the root bark, 7-methyljuglone and isodiospyrin were isolated. Both substances were characterized by comparison with reference compounds (MP; UV; EI-MS; <sup>1</sup>H-NMR; HPLC). Mono- and bidesmosidic saponins of the methanolic extract were isolated by different chromatographic methods, including droplet counter-current chromatography (DCCC)[1] with n-BuOH-acetone-H<sub>2</sub>O 33:10:50 (descending). The structures of the saponins were established by acidic and basic hydrolyses, FAB-MS, <sup>1</sup>H-NMR and <sup>13</sup>C-NMR spectroscopy as derivatives of the triterpene oleanolic acid. Further apolar extracts of Diospyros species (D. usambarensis, D. whyteana, D. lycioides) were analyzed by on-line HPLC-UV spectroscopy using a photodiode array detector. This method gave very rapid information (UV-spectra, retention times proportions) about naphthoquinones in the µg range and is very useful for the screening of naphthoquinone-containing plants. 7-Methyljuglone and isodiospyrin show strong fungicidal (Cladosporium cucumerinum) and molluscicidal (Biomphalaria glabrata) activities [2]. A cytotoxic activity of isodiospyrin in human colon cancer cell lines was shown. The monodesmosidic saponin was strongly molluscicidal against the schistosomiasis-transmitting snail Biomphalaria glabrata.

[1] K. Hostettmann, M. Hostettmann and A. Marston, Nat. Prod. Reports 1, 471 (1984).

[2] A. Marston, J.D. Msonthi and K. Hostettmann, Planta Med. 50, 279 (1984).



UNCINATONE, A NEW ANTIFUNGAL DITERPENOID HYDROQUINONE  
FROM CLERODENDRUM UNCINATUM

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Several species of the genus Clerodendrum find use in African traditional medicine (1-2). The roots of one of these, Clerodendrum uncinatum, have a reputation among traditional healers as cure for bilharzia (3). Phytochemical investigation of this plant has led to the isolation of a compound strongly fungitoxic against Cladosporium cucumerinum. Flash chromatography on silica gel of the petroleum ether extract of Clerodendrum uncinatum followed by crystallization yielded the new hydroquinone diterpenoid uncinatone. The structure of uncinatone was established by spectroscopic methods and X-ray analysis.

This is only the second time that a diterpenoid hydroquinone has been isolated from the family of Verbenaceae, the first report being the isolation of royleanone and dehydroroyleanone from Clerodendrum inerme. Uncinatone inhibited the growth of the spores of Cladosporium cucumerinum at a minimum concentration of 0.5 µg in a TLC assay (4). Other tests with various microorganisms are underway. Furthermore, to confirm indications from traditional healers using infusions of Clerodendrum uncinatum in the cure of schistosomiasis, tests are in progress to evaluate the possible effects of Clerodendrum uncinatum against schistosomes, parasites found in humans suffering from bilharzia.

- (1) F. Haerdi, Afrikanische Heilpflanzen in Acta Tropica suppl. 8, Verlag Recht und Gesellschaft Basel, (1964) 151-2.
- (2) J. Kerharo & J.G. Adam, La pharmacopée sénégalaise traditionnelle, éd. Vigot Frères Paris, (1974) 774.
- (3) J. Williamson, Useful Plants of Malaŵi, The University of Malaŵi, Government Printer, Zomba, (1975) 73.
- (4) D.A. Smith, Phytoalexins, J.A. Bailey, J.A. Mansfield Eds, Glasgow and London, (1982) 220.

132 | BIOLOGICAL ACTIVITY OF LEOCARPOSIDE FROM  
SOLIDAGO L.

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Leiocarposide, a 2'- $\beta$ -D-glucopyranosyloxybenzyl-4- $\beta$ -D-glucopyranosyloxy-2-hydroxy-3-methoxybenzoate, has been isolated for the first time from Solidago virgaurea L. var. leiocarpa /Benth./A.Gray /1,2/ and was found in S. virgaurea L. /3,4/. This compound was isolated in pure state on a gram scale and its diuretic properties compared to that of furosemide were measured in male, white rats /5/. The glucoside appeared to be less toxic - LD<sub>50</sub> = 1.55 g/kg/ furosemide LD<sub>50</sub> = 0.80 g/kg/ and in a dose 25 mg/kg exerted 15 % -20 % weaker action after intraperitoneal /ip/ and peroral /po/ administration than furosemide in a dose 6 mg/kg. The action of leiocarposide was 30 % higher after ip administration than after po use. Although its diuretic active dose = 1/60 LD<sub>50</sub> was lower than that of furosemide, active dose = 1/130 LD<sub>50</sub>, the diuresis caused was delayed /after 5 hs/ and longer lasting /up to 24 hs/. The leiocarposide showed no synergism with flavonoid and saponoside fractions isolated from the same plant material. The glucoside and fractions studied did not influence the Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>2+</sup> excretion in urine. Recent investigations showed the urolithiasis-limiting action of leiocarposide. The 2,4-dihydroxy-3-methoxybenzoic acid obtained on hydrolysis of leiocarposide and identified by IR, MS and <sup>1</sup>H-NMR of its diacetate, showed no diuretic activity.

- 1 K.Hiller, R.Gil-Rjong, P.Franke, E.Gründemann, Pharmazie, 34 /1979/ 360.
- 2 E.Gründemann, R.Gil-Rjong, K.Hiller, Pharmazie, 34 /1979/ 430.
- 3 L.Skrzypczak, Ellnain-Wojtaszek, Pol.J.Chem. 55 /1981/ 683
- 4 L.Skrzypczak, G.Nowak, M.Ellnain-Wojtaszek, Acta Polon. Pharm. 40 /1983/ 637.
- 5 A.Chodera et al. Acta Polon. Pharm. 42 /1985/ 199.

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Isolated individual components of herbal medicines often do not demonstrate the efficacy of the original materials. One explanation which has been under-investigated is the possibility of the formation of stable complexes in situ with modified biopharmaceutical properties. In Chinese herbal medicine polypharmacy is endemic with 4-20 different plant materials being included in each preparation. Noguchi et al (1,2) have noted that Glycyrrhiza is a common component and have shown that glycyrrhizin forms complexes with berberine which enhance absorption. They have claimed that the basis of the complex is ionic interaction between the glucuronic acid carboxyls and the quaternary centre on berberine.

Glucuronides are apparently rare in plants but the flavonoid glucuronide baicalin occurs in Scutellaria baicalensis, another common feature of Chinese herbal preparations. We have shown that this also produces similar complexes with berberine but it appears that glucuronic acid moieties are not essential as we have also demonstrated combination with the aglycone of glycyrrhizin (glycyrrhetic acid) and the flavonoid glycoside rutin. It is therefore clear that the extent of possible interactions is much wider than previously indicated. Bile acids are thought to be important in the absorption of certain cations and we have observed that some of these can also produce similar complexes with berberine.

A range of complexes have been isolated and partially characterised by UV, IR, HPLC and partition behaviour and their practical significance considered.

- (1) Mamoru Noguchi, Chem.Pharm.Bull., 26(9) (1978) 2624-2629
- (2) Mamoru Noguchi, Michinori Kubo, Teruaki Hayashi & Meizi Ono, Chem.Pharm.Bull., 26(12) (1978) 3652-3657



132 c | STRUKTURUNTERSUCHUNG DER SCHLEIMPOLYSACCHARIDE AUS VER-  
BASCUM UND SOLIDAGO UND TESTUNG IHRER ANTITUMORWIRKUNG.  
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In Verbindung mit dem Screening nach potentiell immunstimulierenden (1) und antitumorwirksamen Polysacchariden (2) wurden die wasserlöslichen Gesamtpolysaccharide aus *Verbasci flos* und *Solidaginis herba* isoliert (3). Eine Reinigung und Fraktionierung dieser Polysaccharide erfolgte an DEAE-Sephacel. Homogenitätsuntersuchungen und Molekulargewichtsbestimmungen der einzelnen Fraktionen wurden mittels Hochspannungselektrophorese und Gelpermeationschromatographie durchgeführt. Die qualitative und quantitative Zuckerzusammensetzung der Polysaccharid-Fractionen wurde durch PC, DC und GLC ermittelt. Die Testung der Antitumoraktivität wurde am subcutan transplantierten Sarkom - 180 der Maus durchgeführt. (4) Die Polysaccharide wurden in Konzentrationen von 5 und 25 mg/kg Körpergewicht, beginnend am Tag 1 nach Tumortransplantation ( $2 \times 10^6$  Tumorzellen/Maus) an 10 aufeinanderfolgenden Tagen intraperitoneal appliziert. Eine neutrale Polysaccharid-Fraktion aus *Solidago canadensis herba* zeigte bei einer Dosis von 5 mg/kg eine hohe Antitumoraktivität. Dabei entsprach bis zum Tag 10 das Tumorstadium der Behandlungsgruppe dem der Kontrollgruppe; erst nach diesem Zeitpunkt wurde die antitumorale Wirkung sichtbar, die nach 30 Tagen zu einer Inhibition von 99 % und einer Regression von 75 % führte.

- (1) H. Wagner  
Immunstimulantien aus Pilzen und Höheren Pflanzen  
Fortschritte in der Arzneimittelforschung 133-148 (1984)
- (2) R. L. Whistler, A. Bushway and P. P. Singh  
Nontoxic Antitumor Polysaccharides  
Adv. Carbohydr. Chem. Biochem. 32, 235 (1976)
- (3) M. Belkin, W. G. Hardy, A. Perrault, H. Sato  
Cancer Research Vol. 19, V 1050-59 (1959)
- (4) K. Tabata, W. Ito, T. Kojima, S. Kawabata and A. Misaki  
Carbohydrate Research, 89, 121-135 (1981)

ZELLFREIE SYNTHESSE DER O-SUCCINYLBENZOE SäURE AUS  
ISO-CHORISMIN SÄURE, DIE SCHLÜSSELREAKTION DER VITAMIN K<sub>2</sub>  
(MENACHINON) BIOSYNTHESE

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Zellfreie Extrakte von Escherichia coli AN 154 wurden durch Ultraschallbehandlung gewonnen; das o-Succinylbenzoesäure-produzierende Enzymsystem wurde mit säulenchromatographischen Methoden (Ionenaustauscher, Molekularsieb, Chromatofokussierung) angereichert. Die qualitative und quantitative Identifizierung der o-Succinylbenzoesäure erfolgte durch HPLC.

In zellfreien Extrakten von Escherichia coli AN 154 konnte die Thiaminpyrophosphat-abhängige Reaktion von 2-Ketoglutar Säure mit Isochorisminsäure, aber nicht mit Chorisminsäure, zu o-Succinylbenzoesäure nachgewiesen werden. Damit wurde die von Meganathan und Bentley (1,2) publizierte Biosynthese der o-Succinylbenzoesäure aus Chorisminsäure eindeutig widerlegt. Die erstmals von Campbell (3) postulierte Additionsverbindung aus Succinsemialdehyd und Thiaminpyrophosphat, welche in Form einer Michael-Addition mit Chorisminsäure reagieren sollte, konnte auf dem Biosyntheseweg zur o-Succinylbenzoesäure erstmals nachgewiesen werden.

Das o-Succinylbenzoesäure-produzierende Enzymsystem aus Escherichia coli AN 154 konnte angereichert und charakterisiert werden.

- (1) R. Meganathan und R. Bentley, J.Bacteriol. 153 (1983) 739
- (2) R. Meganathan, J.Biol.Chem. 256 (1981) 9386
- (3) I.A. Campbell, Tetrahedron Letters 54 (1969) 4777
- (4) A. Weische und E. Leistner, Tetrahedron Letters 26 (1985) 1487

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Die arzneilich genutzte (1) Euphorbiacee *Acalypha indica* enthält das cyanogene 3-Cyano-6-pyridonglucosid Acalyphin (2). Bei Verletzung der Pflanze wird massiv Blausäure freigesetzt, was auf das Vorliegen entsprechender  $\beta$ -Glucosidasen hinweist. Fraktionierte Ammonsulfat-Fällung eines hypotonen Blatthomogenats ergab eine Rohenzymfraktion, die DISK-elektrophoretisch vier (A-D) gegen Me-Umbelliferylglucosid aktive Zonen lieferte. Jede der Glucosidase-Banden spaltet sowohl Acalyphin als auch p-Nitrophenyl- $\beta$ -D-glucosid im selben Verhältnis. Zone A repräsentiert 90% der Aktivität; sie besitzt ein MG von 40.000 D; alle anderen Zonen stellen Mehrfache von 40.000 dar (B=80.000; C=160.000; D=320.000). Bande B ist in Bande A konvertierbar. Damit handelt es sich wahrscheinlich um ein in multiplen Formen vorliegendes Enzym. pH-Optimum (5,2) und Temperaturoptimum (50°C) liegen im Bereich anderer  $\beta$ -Glucosidasen (3). Umsatzgeschwindigkeiten (1 mM Substrat) und  $k_M$ -Werte für verschiedene Substrate betragen:

	$k_M$ mmol/l	rel. Aktivität $\mu\text{mol/min} \cdot \text{mg Prot.}$	%
a) p-Nitrophenyl- $\beta$ -D-glucosid	0,20	45,3	100,0
b) p-Nitrophenyl- $\beta$ -D-galaktosid	0,38	42,5	93,8
c) p-Nitrophenyl- $\alpha$ -D-glucosid	-	0,22	0,5
d) Acalyphin	0,43	75,4	166,4
e) Prunasin	0,75	28,3	62,4
f) Amygdalin	-	0	0
g) Linamarin	-	0,31	0,7
h) Coniferin	0,38	10,5	23,2

Zwar wird Acalyphin am besten umgesetzt, jedoch werden auch andere Substrate (insb. b,e,h) gut hydrolysiert. Damit unterscheidet sich das vorliegende Enzym von beschriebenen hochspezifischen  $\beta$ -Glucosidasen (3). Wahrscheinlich hat es neben der prämortalen HCN - Produktion weitere physiologische Aufgaben (z.B. bei der Lignifizierung).

1. Hager's Handbuch der Pharmazeutischen Praxis, Bd.II, p.876, Springer Verlag, Berlin, Heidelberg, New York (1979)
2. A. Nahrstedt, J.-D. Kant, V. Wray, *Phytochemistry* **21** (1982) p. 101-105
3. W. Hösel, The Enzymatic Hydrolysis of Cyanogenic Glucosides, in: B. Vennesland et al. (Eds.), *Cyanide in Biology*, Academic Press, London (1981), p. 217



<sup>14</sup>C - INKORPORATION IN DIE FLAVONOLE DER BLÄTTER  
VON GINKGO BILOBA

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Extrakte aus den Blättern von *Ginkgo biloba* L. werden erfolgreich bei peripheren und cerebralen Durchblutungsstörungen eingesetzt (1), (2). An der Wirkung sollen Flavonolglykoside maßgeblich beteiligt sein. Kenntnisse über Resorption, Verteilung und Verweildauer in Organen liegen bisher nicht vor. Um Grundlagen dafür zu schaffen, wurden Versuche zur Radioisotopenmarkierung mit dem Ziel durchgeführt, den Aglykonteil spezifisch hoch zu markieren. Als Precursoren wurden [<sup>14</sup>C]Natriumacetat und L-[U-<sup>14</sup>C]Phenylalanin an junge G.b. Langtriebe appliziert. Zur Optimierung der Precursorinkorporation wurde der Einfluß von Beleuchtungsstärke und Inkubationszeit auf die Einbauraten untersucht. Die Aglykone wurden nach Hydrolyse des Methanolextraktes isoliert und deren spezifische Aktivität durch DC/LSC und HPLC bestimmt.

Die Einbauraten von markiertem Phenylalanin bzw. Acetat stiegen während einer 32-tägigen Versuchsdauer bei einer Beleuchtungsstärke von 12.000 Lux kontinuierlich an und erreichten schließlich 0,3 % bzw. 0,05 %. Die Beleuchtungsstärke beeinflusste die Einbauraten sehr stark, mit 1.000 Lux wurden nur noch 0,05 % bzw. 0,01 % der applizierten Aktivität eingebaut. Die zusätzliche Gabe von unmarkiertem Phenylalanin steigerte die Einbauraten nicht. Die spezifische Aktivität der Aglykone nahm während der Versuchsdauer stark ab. Die Einflußfaktoren wirkten sich bei den drei Hauptaglykonen Quercetin, Iso-rhamnetin und Kämpferol in gleichem Maße aus.

Flavonole mit einer spezifischen Aktivität von 0,015 - 0,02 mCi/mmol lassen sich durch Gabe von L-[U-<sup>14</sup>C]Phenylalanin bei hoher Beleuchtungsstärke und nach einer Inkubationszeit von 2-4 Tagen gewinnen.

- (1) Peter, H.-J., Fisel, J. et al: *Arzneim.-Forsch.* 16, 719 - 725, (1966).
- (2) Le Poncin-Laffitte, M., Rapin, J. et al: *Arch. Int. Pharmacodyn. Ther.* 243, 236 - 244, (1980).

# 138 Vitamin K<sub>2</sub>-Biosynthese — Strukturaufklärung, Biosynthese und Funktion der OSB-CoA-Ester

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Das erste aromatische Intermediat des Vitamin K<sub>2</sub>-Biosyntheseweges, die ortho-Succinylbenzoesäure (OSB), wird durch zellfreie Extrakte von Mycobacterium phlei und Escherichia coli ATP- und CoA-abhängig umgesetzt zur 1,4-Dihydroxy-2-naphthoesäure (DHNS). Vorausgehende Versuche (1) hatten gezeigt, daß ein OSB-CoA-Ester an dieser Ring-schlußreaktion beteiligt ist.

Da die OSB eine Dicarbonsäure ist, galt es nun alle drei möglichen Strukturvarianten dieses OSB-CoA-Esters chemisch zu synthetisieren. Dies gelang über die energiereichen Imidazolyl-Derivate der OSB als Zwischenstufen, die dann exergonisch mit CoASH zu den korrespondierenden OSB-CoA-Estern weiterreagierten. Die Trennung dieser extrem labilen Verbindungen gelang mittels HPLC an RP-8; UV- und FAB-Massenspektren dienten zur Identifizierung (2).

Überraschenderweise konnte auch in zellfreien Extrakten von M. phlei die enzymatische Bildung aller drei möglichen OSB-CoA-Ester bei pH 7,9 nachgewiesen werden. Inkubiert man jedoch bei pH 6,5, wird lediglich die aliphat. -COOH der Succinylseitenkette der OSB mit CoA aktiviert.

Versuche mit <sup>14</sup>C-markierten Substraten ergaben, daß nur dieser "aliphatische" OSB-CoA-Ester enzymatisch zum Folgeprodukt DHNS umgesetzt wird; die beiden anderen Strukturvarianten sind physiologisch inaktiv (2), (3).

(1) Heide, L., S. Arendt, and E. Leistner, J. Biol. Chem. 257, (1982) 7396

(2) Kolkmann, R., and E. Leistner, Tetrahedron Lett. 26 (1985) 1703

(3) Kolkmann, R., Dissertation Bonn 1985

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Nach bisherigen in vivo Versuchen (1) ist über die Bildung von Flavon-C-Glycosiden bekannt, dass die C-glycosidische Bindung nicht am Flavon, sondern vermutlich an Vorstufen wie Chalcon oder Flavanon erfolgen muss.

Als Modell für die Biosyntheseuntersuchungen wurden Keimlinge von Fagopyrum esculentum gewählt, die in den Kotyledonen aus Reservestoffen (2) die Flavon-C-Glycoside Vitexin, Isovitexin, Orientin und Isoorientin bilden. Die Bildungskinetik wurde unter verschiedenen Bedingungen untersucht. Um weitere Erkenntnisse über die Biosynthese und mögliche Zwischenstufen zu gewinnen, wurden Enzymextrakte der Kotyledonen in vitro unter verschiedenen Bedingungen mit den Substraten Naringenin, bzw. 2',4,4',6' Tetrahydroxychalcon und UDP[<sup>14</sup>C]-Glucose inkubiert. Die Bestimmung der Produkte erfolgte durch Ausschütteln, DC (Kieselgel) und quantitative HPLC (RP 18).

(1) M. Grün und G. Franz  
Planta Medica 47 (1983) 131

(2) U. Margna und T. Vainjäär  
Z. Naturforsch. 38c (1983) 711



AMYLOID (XYLOGLUCAN) FORMATION IN THE COTYLEDONS OF  
TROPAEOLUM MAJUS SEEDS

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Mature seeds of *Tropaeolum majus* L. contain the cell wall polysaccharide xyloglucan (amyloid), protein and lipid as storage substances (1). The transitory occurrence of starch during the process of seed development could be substantiated. The different stages of seed ripening are demonstrated by light- and electron micrographs.

[U-<sup>14</sup>C]-labelled xylose, glucose and glucuronic acid were fed to ripening seeds and the incorporation of radioactivity into xyloglucan, starch and the sugar nucleotide fraction of the amyloid forming cotyledons was determined.

The results indicate that exogenous supplied xylose is not incorporated directly into xyloglucan, but can be utilized only after a transformation to glucose. Radioactivity from glucuronic acid was predominantly found in the xylose moiety of xyloglucan.

Incubation of seeds with [6-<sup>14</sup>C]-labelled glucose resulted in a specific incorporation of glucose into the amyloid. Xylose residues of the amyloid remained unlabelled, demonstrating a direct metabolism of UDP-glucose to UDP-glucuronic acid. These findings were substantiated by the occurrence of the respective NDP-sugars which are known to be the precursors for polysaccharide synthesis.

- (1) R. Hegnauer, Chemotaxonomie der Pflanzen, Band 6, Birkhäuser-Verlag,  
Basel und Stuttgart, 1973, p.538.

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